A study of the phytoplankton of Swansea Bay.

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A STUDY OF THE PHYTOPLANKTON OF SWANSEA BAY

A thesis submitted to the University of Wales in candidature

for the degree of Philosophiae Doctor

by

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(B.Sc. Alexandria)

April 1986

To my parents, my wife, and my daughter

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ACKNOWLEDGEMENTS

I would like to express my gratitude to my supervisor Dr J.H. Hayward for his help, advice and encouragement throughout the course of this study. My special gratitude goes to the University College of Bahrain for its generous grant. I would also like to thank Professor P.J. Syrett for his hospitality and for providing the facilities to carry out this work. My thanks to Dr S.J. Wainwright for his assistance in the statistical analysis of the data and to Mrs E. Griffiths for providing the deionized water, and for all her help. Many thanks to Captain D.B. Davies and all the crew of the R.V. "Ocean Crest" for their help and friendship during the fieldwork. My deep gratitude to my wife for her love, support, encouragement and patience, especially during the hard times.

Finally, I would like to thank Mrs G. Bridges and Mrs M. Owens for typing this thesis.

ABSTRACT

The intention at the beginning of this study was to investigate the primary productivity of phytoplankton in Swansea Bay. This productivity was measured fortnightly using the ^{14}C-fixation technique. Biomass standing stock was measured as chlorophyll *a* m^{-3} and as cell concentration. Concurrently, several environmental factors were measured to relate any possible effect on phytoplankton growth and species succession. The factors studied were seawater temperature, salinity, nitrate, nitrite, ammonia, phosphate, silicate and some meteorological data supplied by a local station. A weak positive correlation was found between net phytoplankton cell number and phytoplankton chlorophyll *a*. Based on this finding, the relative importance of phytoplankton size classes as primary producers was investigated. As a result of this investigation, it has been found that most of the primary productivity (80%) was carried out by phytoplankton less than 20 µm. Special attention was given to the role of nitrate in the ^{14}C-fixation by different phytoplankton size classes and by phytoplankton species grown in synthetic medium. The effect of nitrate on the ^{14}C-fixation by the size classes was investigated using nutrient enrichment technique. From these experiments it has been found that nitrate enrichment stimulates the increase of ^{14}C-fixation by the different size fractions at different rates. When nitrate was given to nitrogen-starved *Thalassiosira sp.* and *Asterionella japonica*, it was found that nitrate was taken up at the expense of ^{14}C-fixation. It was also found that algal cells cultured in medium with high nitrate concentration increased their chlorophyll *a* content. It was concluded from this study that nano and picoplankton play an important role in the productivity of Swansea Bay, and that nitrate plays a significant role as a limiting nutrient not only to the primary productivity of the phytoplankton population as a whole but also to the different phytoplankton size classes.

CHAPTER I

GENERAL INTRODUCTION

In the many years spent in the study of marine biology, plankton research has received increasing attention. Phytoplankton studies began with the observation of Joseph Hooker in 1839 that the green water and slimy brownish-green scum on the Antartic ice were plant-like. Victor Hensen in 1887 adopted a more physiological approach to the study of phytoplankton. The first review of the observations in the early years of phytoplankton studies was conducted by Gran (1912).

Phytoplankton are the major contributors to primary productivity in the oceanic environment. Apart from seaweeds and higher plants, phytoplankton are the main producers in coastal environment. Their role as major primary producers of organic material in the aquatic envrionment made them the core of primary productivity studies. Primary productivity is defined as the amount of carbon fixed by autotrophic organisms through the synthesis of organic matter from inorganic compounds such as CO_2 and H_2O using energy derived from solar radiation or chemical reactions. The total amount of organic matter produced by photosynthesis represents the gross primary production. Net primary production represents the amount of organic matter remaining from gross primary production after the deduction of the amount of organic matter used in cell maintenance through respiration. Different methods are used to measure net primary production. The oxygen method of Gran has been used since the early decades of the twentieth century. Another method was developed by Steeman Nielsen (1952). This new method involves the measurement of carbon fixed by phytoplankton using carbon isotope (^{14}C). Since there is a direct relationship between the amount of chlorophyll _a_ in a phytoplankton sample from a given volume of water and gross primary productivity, chlorophyll _a_ measurement has been used as an indirect tool in the

measurement of primary productivity. One of the most important features of phytoplankton population dynamics is species succession. This phenomenon was observed by many authors (Marshall and Orr, 1927; Lillick, 1940; Robinson, 1965; Hulbert, 1975; Butler et al., 1979). In the Bristol Channel, species succession has been reported a number of times (Rees, 1939; Pearce, 1967; Paulraj, 1974; Tyler, 1976; Vogelmann, 1980; Sexton, 1985). Since phytoplankton vary in size from 2 to 300 μm, this means that it is likely for a phytoplankton population of specific cell volume to be replaced by another population with a different cell volume.

The diversity of cell size leads to the introduction of phytoplankton differentiation into different groups according to their size. The term nanoplankton was first used by Lohmann (1903) for phytoplankton not retained by phytoplankton net. Sieburth et al. (1978) used the term picoplankton to describe plankton smaller than 2 μm in diameter.

Primary productivity by phytoplankton size fractions was investigated in the different environmental ecosystems (McAllister et al., 1959; Malone, 1971a; Durbin et al., 1975). In general, it has been observed that nano and picoplankton dominate the oceanic environment, while the net plankton dominate the coastal and neritic environment (Malone, 1980). Exceptions to these observations were found (Durbin et al., 1975; Hannah and Boney, 1983).

Many factors have been found to influence or limit primary production and the distribution of phytoplankton in time and space. These factors include light (Marshall and Orr, 1927, 1930; Thomas et al., 1978; Chan, 1980), temperature (Riley, 1946; Fogg, 1975; Raymont, 1980; Harrison and Turpin, 1982), nutrients (Atkins, 1930; Riley, 1937; Ketchum et al., 1958; Ryther and Kramer, 1961; Hulbert, 1970; Pingree et

al., 1977), and grazing (Bainbridge, 1953; Mullin, 1963; Martin, 1970).

The effects of the different environmental factors on the growth, species succession and physiology of phytoplankton have been studied intensively in the natural environment and in cultures (Fogg, 1975; Raymont, 1980; Morris, 1980).

Cultures have been used as simulated environments to simplify the complexity of the natural environment. In respect to the effect of environment factors (especially nutrients) on the growth of phytoplankton in unialgal, mixed cultures, or even natural populations, bioassay technique has been widely used (Schelske and Stoermer, 1971; Schindler, 1971; Goldman, 1972; Barlow et al., 1973).

The present research started with a field study of the primary production, species succession, size fractionation, and the seasonal change in the physical and chemical environmental factors. Concurrently, culture experiments were carried out to investigate the relationship between phytoplankton growth expressed either as ^{14}C-fixation or cell concentration and nutrient enrichment, especially nitrate.

Throughout this thesis, the presentation of the chapters follows the development of the results and the findings based upon them. The chapters are presented in the following order:

Chapter II — Describes the study area, its location, hydrography, and the methodology used in the field studies.

Chapter III — Describes and discusses the seasonal change in the physical and chemical factors: sunshine hours, sea and water temperature, rainfall; concentrations of nitrate, nitrite, ammonia, phosphate and silicate.

Chapter IV Describes and discusses the seasonal change of phytoplankton abundance, biomass, primary productivity as well as the zooplankton abundance.

Chapter V Describes and discusses phytoplankton species succession.

Chapter VI Describes and discusses the seasonal change in the primary productivity of phytoplankton size classes.

Chapter VII Describes the methodology used in the laboratory studies which were (among other experiments) devoted to study the response of phytoplankton, in cultures or natural seawater, to the addition of nutrients (mainly nitrate).

Chapter VIII The findings of the research are discussed, connected and final conclusions are drawn.

CHAPTER II

STUDY AREA AND METHODOLOGY

STUDY AREA

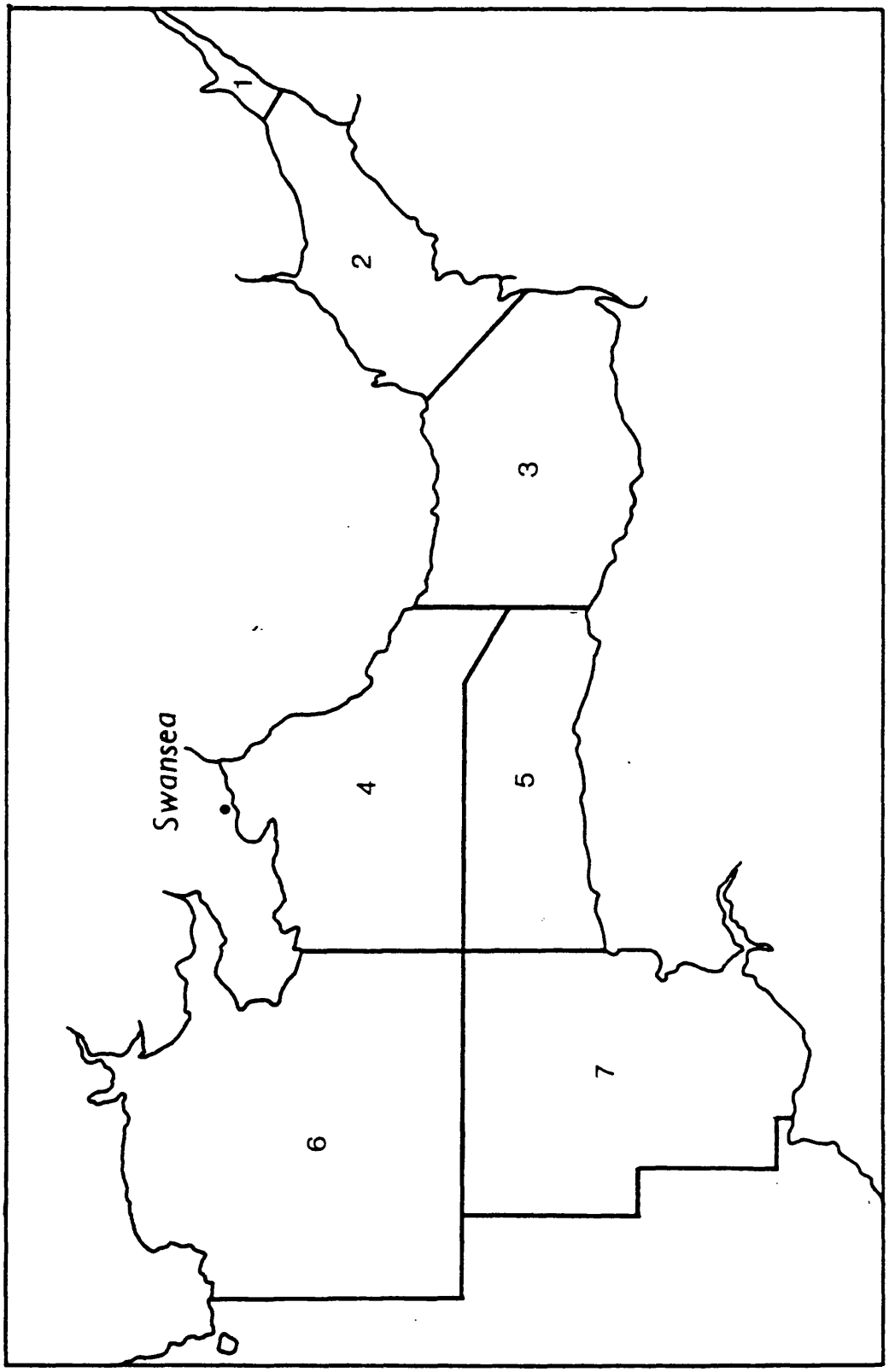
The Bristol Channel is one of the main water environments in the United Kingdom. Its significance derives from the presence of many industrial and economic sites along its northern and southern coastlines. The Severn Estuary and Bristol Channel form a wide, shallow estuary. Rivers Severn, Avon, Usk, Wye, Taff, Parret, Axe, Taw, Torridge, Neath and Tawe form the main run-off sources into the Bristol Channel. The mean annual freshwater flow into the estuary is 8×10^9 m^3 $year^{-1}$ (Joint and Pomroy, 1981). The water circulation in the Bristol Channel consists mainly of an oscillatory tidal current and a relatively small residual current (Uncles, 1984). The ecology and hydrodynamics of the Bristol Channel are affected directly by the tidal and residual currents. They produce a frictional drag on the sea bed. The large tidal stresses produced are associated with strong vertical current shear which generates intense vertical mixing, which is responsible for the high turbidity of the channel.

Based on ecological and oceanographical features, the Bristol Channel is divided into seven hydrodynamic regions (I.M.E.R., 1974, 1975) (Fig. 2.1). Tidal currents in the central regions of the channel are mainly linear and parallel to the northern and southern coastlines (Admiralty Chart No. 1165, Admiralty Tidal Stream Atlas, 1973). Modifications to this rectilinear system occur along the South Gower coastline, due to the presence of embayments. Examples of such cases are the eddy systems in Port Eynon and Oxwich Bays (Tyler and Banner, 1977; Ferentinos and Collins, 1980).

Swansea Bay is one of the largest embayments along the South Gower coastline at the northern boundary of the Bristol Channel. The

Figure 2.1 Hydrodynamic regions of the Bristol Channel

1. Inner estuary
2. Outer estuary
3. Inner channel
4. North central channel
5. South central channel
6. North outer channel
7. South outer channel


1
2
3
4
5
6
7
Swansea

distance, at the entrance of the bay, between Mumbles Head in the west to the eastern limits of the bay is about 12 km, and the distance from this boundary to the northern shore line is about 4 km (Collins et al., 1979) (Fig. 2.2). The average depth of the bay is 8 m.

Many rivers, streams, domestic and industrial discharges contribute to the input into Swansea Bay (Fig. 2.2). The mean volumetric input to the bay from rivers and discharges during a sampling programme conducted by Chubb et al. (1980) was 37.9 $m^3.sec^{-1}$. Rivers and streams accounted for 93.6% of this flow and the two major rivers, Tawe and Neath together contributed 66.8% of the total.

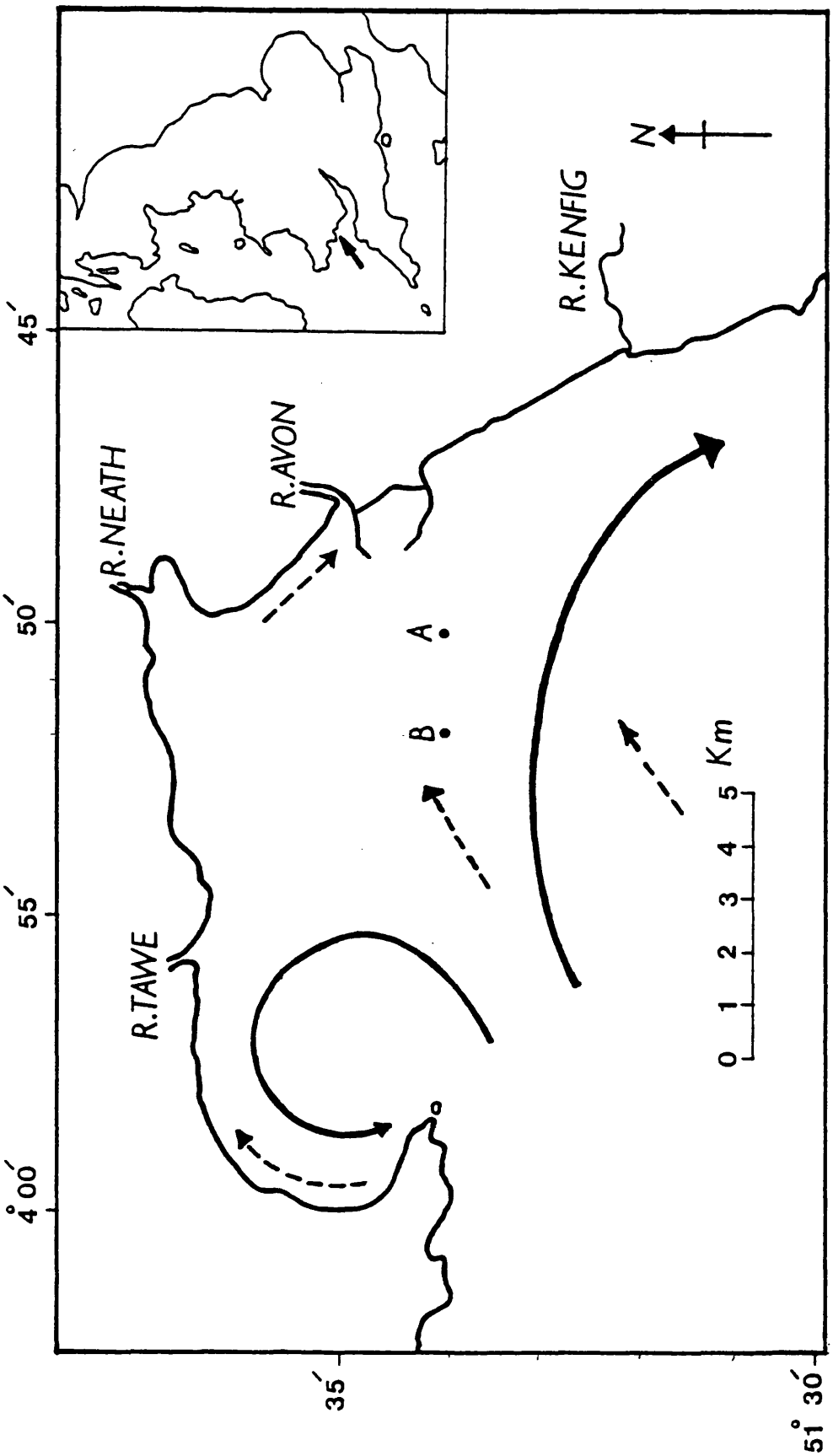
Due to the protrusion of headlands in the bay, the rectilinear tidal current of the central region of the Bristol Channel is modified to an anticlockwise eddy circulation pattern in the west region of Swansea Bay. In the eastern part of the bay, an area of divergence between the main rectilinear current and the anticlockwise eddy circulation was observed (Collins et al., 1979) (Fig. 2.2).

The configuration of the coastal lines due to the presence of heads and land intrusions affected the quality and the hydrodynamics of the water by confining the mixing between offshore water and the inshore water to the inner bay. This was explained in the results of the work carried out by Mantoura and Morris (1980). They found a water mass representing the later stages of mixing between the coastal waters of inner Swansea Bay, with the main offshore water of the Bristol Channel. This is confirmed by the presence of an area with low salinity water during the summer months near Mumbles Head (Joint, 1980).

Figure 2.2 Swansea Bay

A: Eastern sampling station
B: Western sampling station

⟶ Tidal transport
- - -▶ Wave transport


R.TAWE
R.NEATH
R.AVON
R.KENFIG
A
B
N
0 1 2 3 4 5 Km
4° 00′
55′
50′
45′
35′
51° 30′

METHODOLOGY

I. Field Collection of Samples

The field work of this study was carried out using RV 'Venturous'. The water and plankton samples obtained were from an area in the eastern region of Swansea Bay. Two sampling stations were established to the east (A), and west (B), of the BP outfall bouy (51° 34'N, 3° 51'W) and at a distance of half a nautical mile from it (Fig. 2.2). It is very likely that the sampling stations are in the area of the rectilinear currents, and near the suggested divergence area at the eastern part of the bay (Ferentinos, 1978; Collins et al., 1979).

The water column in Swansea Bay is usually well mixed due to the presence of strong tidal excursions. For this reason the water samples were pumped from one depth only (1 foot below the surface).

To obtain the best results, the same water mass should be sampled each time in order to observe and compare changes occurring in the water quality. In the case of Swansea Bay, this is difficult due to the complex water hydrodynamics and the powerful tidal action characteristic of Swansea Bay, and the Bristol Channel as a whole.

This means that, when, at a fixed station, water is sampled at different times during the tidal cycle and at different intervals during the lunar cycle, the water collected could be representing different water masses (Vogelmann, 1980).

Taking these points into consideration, a programme was established to collect water and plankton samples at each spring tide, and within 2 hours from high tides for a period of 28 months.

i. Plankton collection

A standard plankton net was used for the plankton collection. The net consisted of an 18" diameter ring. To this ring, a cone of bolting silk with a mesh size no. 155 was attached. The end of the cone was open so that a jar was tied to it during the towing. Starting from the bouy, the net was towed first to the east for 20 minutes, and then to the west for another 20 minutes, and at each tow a line of about half a nautical mile was covered.

The plankton collected in the sampling jar were poured gently into plastic bottles.

ii. Water sampling

Seawater samples were collected from each station using a tap fixed on board the ship. The water was collected in clean, 5 litre capacity, polythene bottles. The water temperature was measured immediately using a -10°C to +110°C mercury thermometer. Because the samples usually returned to the laboratory within 1 hour from sampling, they were kept in the shade until reaching the laboratory where the treatment was carried out.

II. Laboratory Treatment of Samples

All of the chemicals used were of the highest purity. They were obtained either from BDH or Sigma Chemical Companies. All of the glassware and equipment not used in the radioactive work were soaked overnight in 5% HCL, then rinsed three times with double distilled water prior to their use. The equipment used in the radioactive work was soaked in tap water containing detergent for two days, then rinsed five times with distilled water and left to dry prior to any radio-

active work.

i. Plankton

After returning the plankton samples to the laboratory, 200 ml of each sample was poured into 250 ml polythene bottle. To each sample Lugol's solution (5 g KI + 2.5 g I_2 in 250 ml H_2O) was added (up to 5 ml/200 ml) The plankton were identified and counted within a few days from sampling. Examination of the plankton samples was carried out using PZO compound microscope. With the aid of a fin pipette, 1 ml of the sample was transferred to Sedwick Rafter chamber, total volume 1 ml; the chamber was divided into 1000 squares. Depending on the concentration of the sample, phytoplankton and zooplankton in at least 50 squares were counted and identified. This process was repeated at least three times for each sample.

ii. Seawater sample

As mentioned previously, 5 litres of seawater were collected from each station. From each sample, 100 ml was used for the determination of the alkalinity, and 100 ml for the isolation of phytoplankton. Depending on the seasonal growth, triplicates of 0.5-1 litres of the seawater were filtered through Whatman GF/C filters using Millipore filtration apparatus. The filter discs were used for chlorophyll a determination.

200 ml of the filtrate was stored frozen until the next day for phosphate analysis. 1 litre of the filtrate was used in the analysis of nitrate, nitrite, ammonia and silicate. About 500 ml of the filtrate was stored in a glass, medical flat bottle to be used in the measurement of salinity. 400 ml of the seawater was used in the

measurement of primary production.

a. Salinity

Salinity was measured using YSI Model 33 S-C-T meter. The instrument was calibrated against different known salinities of synthetic seawater (25-35‰). Samples were allowed to equilibrate to room temperature overnight prior to analysis.

b. Chlorophyll a

Chlorophyll a was determined by the method described by Strickland and Parsons (1972). 0.5-1 litre of seawater was filtered through a Whatman GF/C filter disc. The last 200 ml was treated with 1 ml of $MgCo_3$ suspension. The filter disc was divided into small portions and placed in 15 ml centrifuge tube to allow easier extraction. 10 ml of 90% acetone was added to the content of the tube, and the whole contents shaken vigorously to dissolve the filter paper. The mouth of the tube was covered with parafilm, and then the whole tube was covered with aluminium foil in order to minimize the effect of light. The tube was placed in the dark at 4°-5°C for 24 hours. At the end of the storage period, an additional 2 ml of 90% acetone was added and the sample was centrifuged at a speed of 2500 rpm. The supernatant was read in a 1 cm cuvette against a 90% acetone blank on CE 272 linear readout ultra-violet spectophotometer, at wavelengths of 750, 665, 645 and 630 nm. To correct for turbidity, the reading at 750 nm was substracted from the readings at the other wavelengths. The extinction values were multiplied by 12 to normalize them to the values expected from 10 cm cuvette and 10 ml of extract.

The concentration of chlorophyll a was calculated according to

Strickland and Parsons (1972) equations:

$$c \text{ (chlorophyll } \underline{a}) = 11.6 \text{ (E665)} - 1.13 \text{ (E645)} - 0.14 \text{ (630)} \quad (1)$$

$$\text{mg chlorophyll } \underline{a} \ \ m^{-3} = \frac{c}{v} \quad (2)$$

where c was the value obtained from equation (1) and v was the volume of filtered seawater in litres.

Capabilities:

If it is practical, up to 10 litres of seawater can be filtered with a lower limit of:

chlorophyll a precision at the 5 µg level

The correct value lies in the range:

Mean of n determinations $\pm$ $0.26/n^{\frac{1}{2}}$ µg chlorophyll a

c. Primary production

Primary production of the photoautotrophic planktonic organisms was determined by the method described by Strickland and Parsons (1972), based on the original radioactive carbon-14 method by Steemann Nielsen (1952).

This method is based on the assumption that a small amount of specific radioactive compound added to a sample of water containing biological material, is assimilated at about the same rate as the corresponding non-labelled compound occurring naturally in the water to be assayed (Vollenweider, 1974).

Dilution and storage of radioactive carbon

The radioactive carbon was obtained as $NaH^{14}4CO_3$ from Amersham in 1 mci batches. Using a syringe, the contents of the ampoule were transferred to 100 ml volumetric flask. The ampoule was rinsed several times by 0.5 N NaOH to ensure that all the radioactive material was transferred to the flask. The volume was diluted to 100 ml with a 0.5 N NaOH.

This dilution gave a pH of about 9. This was carried out to minimize the loss of ^{14}C during storage and handling (Gargas, 1975). When 1 mci was diluted to 100 ml, the activity obtained was 10 µci/ml. 5 ml portions were distributed into tightly stoppered, 5 ml plastic containers. These working solutions were kept frozen until required.

Determination of the ambient CO_2

The method used here and described by Strickland and Parsons (1972), is based on mixing a known volume of seawater with a standard acid. The initial pH of the seawater was determined immediately after bringing the seawater to the laboratory using CORNING pH meter model 7.

25 ml of 0.01 N HCl was pipetted into a 200 ml, wide mouth bottle. To this acid, 100 ml of the seawater was added. The contents were mixed together and the pH was measured. By measuring the pH before and after the addition of the acid, the total CO_2 was calculated using the given tables (Strickland and Parsons, 1972).

^{14}C uptake

125 ml bottles were used for the artificial light incubator. The incubator used consists of a glass container holding up to 30 litres of water. The incubation bottles were covered with water in this container. The water temperature in the container was controlled using

a Churchill Chiller Thermo-circulator. The temperature of the water was checked using a mercury thermometer. Osram Liteguard Daylight fluorescent tubes were used as a source of light with intensity of 150 $\mu E.m^{-2}$ sec^{-1}. For each station, three light bottles and one dark bottle were filled with 100 ml seawater. The dark bottle was covered by aluminium foil. The dark bottle was used to correct for the dark fixation and respiration. The dark fixation is due to biological and non-biological mechanisms. Biological mechanisms are associated with the tricarboxylic cycle. Non-biological mechanisms are related to adsorption, contamination and to background (Nielsen and Bresta, 1984).

Taking the zero time into consideration, 2 µci was added to each bottle. After 4 hours of incubation, a volume of the sample (20-100 ml) was filtered through 25 mm AA Millipore filter. The suction/pressure used was less than 0.3 $Kp.cm^{-2}$. The filter disc was then exposed to fumes of concentrated hydrochloric acid for 1 minute in order to remove possible extracellular ^{14}C. It was then left in an opened glass vial for 2 hours to allow the escape of any CO_2 formed, prior to the addition of scintillation liquid. The scintillation liquid used was obtained from Amersham. It was called Filter-Count, and was recommended by the manufacturer for productivity studies. 10 ml of this liquid was added to the sample vial, mixed well with the filter disc, and kept for 30 minutes prior to counting to complete the dissolution of the filter disc. The background count and the activity of the added ^{14}C were determined each time the primary production measurement was conducted. Beckman LS 6800 scintillation counter was used. There was no need to calculate the efficiency of the counter because it was calculated internally by the machine and the results were expressed as disintegration per minute in the output.

The total carbon uptake, Pt, during the time, t, was calculated for every sample according to the following equation (Nielsen and Bresta, 1984):

$$Pt = \frac{dpm(a).\text{total } CO_2(c).1.05(d).1.06(e).K_1.K_2}{dpm(b)}$$

where:

Pt = carbon uptake, mg C m^{-3} hr^{-1}
(a) = light dmp-dark dpm = net dpm/sample
(b) = the activity of the added ^{14}C solution, dpm
(c) = concentration of total CO_2 in the experimental water, mg C m^{-3}
(d) = a correction for the effect of ^{14}C discrimination, the uptake of the ^{14}C is 5% slower than that of the ^{12}C
(e) = a correction for the respiration of organic matter produced during the experiment. This has been found to represent 6% at optimal photosynthesis.

K_1 = a correction factor for subsampling
K_2 = a time correction factor

Capabilities:

Range: 0.05-100 mg C m^{-3} hr^{-1}

Precision at the 25 mg C m^{-3} hr^{-1} level

The correct value lies in the range:

Mean of $\underline{n}$ determinations $\pm$ $3/n^{\frac{1}{2}}$ mg C m^{-3} hr^{-1}

(5 hour incubation, 1 µci added)

d. Nutrients

Synthetic seawater

Synthetic seawater was prepared for the standard solution by dissolving 31 g Na Cl, 10 g Mg $SO_4 7H_2O$ and 0.05 g Na $HCO_3.H_2O$ in 1 litre of distilled water.

Nitrate-nitrogen

The determination of nitrate-nitrogen was based on the method described by Wood et al. (1967), the method based on the quantitative reduction of nitrate to nitrite. The sample was treated with tetrasodium ethylenediaminetetraacetate and passed through a column of copperized cadmium filings. The nitrate was reduced to nitrite which was determined by a diazotization method.

Apparatus

The reduction column used was identical to that described by Wood et al. (1967) (Fig. 2.3). It consisted of a U-shaped glass tube. One side of it consisted of a 34 cm long tube, with an inner diameter of 8 mm. At the top of this tube was a 30 mm diameter reservoir holding up to 50 ml. The other arm was 2 mm capillary tubing, curved at the upper part, ending with a Teflon stopcock equipped with a valve. A length of about 10 cm between the bottom of the reservoir and the discharge tip was used to maintain a nearly uniform flow into a graduated cylinder.

Reagents

Double-distilled water was used for the preparation of the reagents.

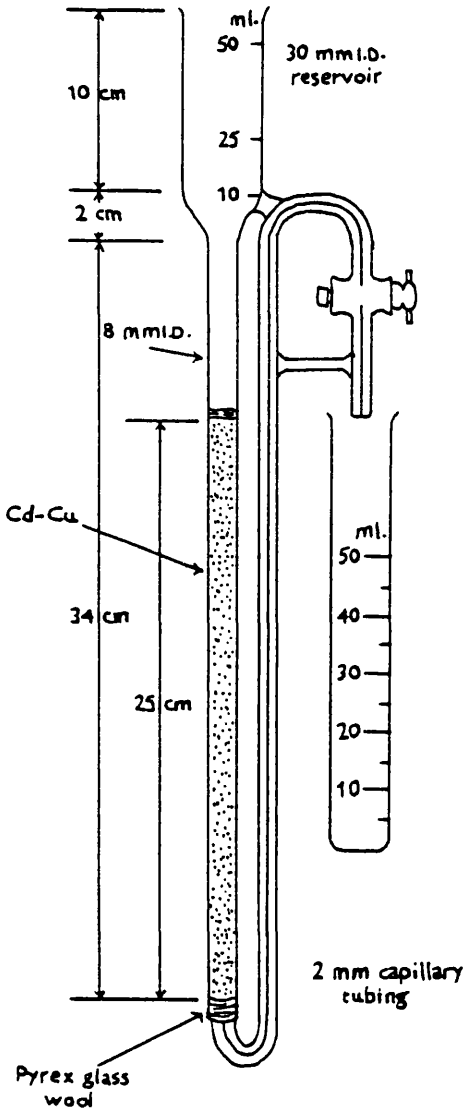
Sulphanilamide solution:

5 g sulphanilamide dissolved in a solution of 50 ml 12 N.HCl and 300 ml of double-distilled water, then diluted to 500 ml with double-distilled water.

N - (1. naphthyl)-ethylendiaminedihydrochloride solution:

0.5 g of the dihydrochloride was dissolved in double-distilled water and diluted to 500 ml.

Figure 2.3 Nitrate reduction column


ml.
50
30 mm I.D.
reservoir
25
10
10 cm
2 cm
8 mm I.D.
Cd-Cu
34 cm
25 cm
ml.
50
40
30
20
10
2 mm capillary
tubing
Pyrex glass
wool

Nitrate standard:

0.1264 g KNO_3 (dried for 1 hour at 110°C) dissolved in and diluted to 250 ml with double distilled water.

Nitrite standard:

0.0863 g $NaNO_2$ (dried for 1 hour at 110°C) dissolved in and diluted to 250 ml with double distilled water.

Copper sulphate solution:

20 g $CuSO_4.5H_2O$ dissolved in 1 litre of double distilled water.

EDTA solution:

38 g of tetrasodium ethylenediaminetetraacetic acid dissolved in and then diluted to 1 litre with double distilled water.

HCl (2N):

85 ml of 12N.HCl diluted to 500 ml with double distilled water.

HNO_3 (0.3N):

10 ml of 15.4 N.HNO_3 diluted to 500 ml with double distilled water.

HCl (0.0015N):

0.125 ml of 12N.HCl diluted to 1 litre with double distilled water.

Column wash solution:

1 ml of the EDTA solution added to 50 ml of 0.0015N.HCl.

Preparation of the column

Sticks of cadmium were filed. The fractions retained by 0.5 mm screen but passing through a 2 mm screen were used. About 40 g of the cadmium filings were washed with 2N.HCl in a separatory funnel, then rinsed thoroughly with distilled water. This was followed by washing with 0.3 N.HNO_3, rinsing with distilled water, and then washing with 2 N.HCl to remove the NO_3 ions. A thorough rinsing with distilled water followed. The cadmium was then treated with 100 ml of the copper sulphate solution in a flask. It was well shaken and then flushed with distilled water, preventing the copperized cadmium from being exposed to air.

A plug of Pyrex glass-wool was placed in the bottom of the tube. The tube then filled with distilled water and the copperized cadmium filings introduced slowly. When a length of 25 cm of filings was reached, a plug of Pyrex glass-wool was placed at the top of the filings.

The filings were washed with approximately 50 ml of the wash solution. Then it was allowed to stand for about 24 hours, renewing the wash solution a few times during that period.

To give about 98% reduction to nitrite, 3 litres of water containing 60 µg, at l^{-1} of NO_3-N and 20 ml of EDTA solution/l were passed through the column.

Seawater sampled and filtered through GF/C membrane filter was used for the nitrate analysis. Nitrate concentration was determined for the water samples, and the reagents. The reduction efficiency of the column was determined by passing water containing a known amount of NO_3^--N and then comparing the resulting quantity of nitrite with that of a water containing a known amount of NO_2^--N.

$$E = \frac{Ac,NO_3^-}{Ac,NO_2^-}$$

E = the efficiency of reduction

Ac,NO_2^- = corrected absorbance of nitrite standard

Ac,NO_3^- = corrected absorbance of a reduced nitrate standard of the same concentration

Procedure

To 50 ml of sample in 50 ml graduated cylinder, 1 ml of EDTA was added. The remains of the previous sample was rinsed with about 10 ml of the sample. When all the 10 ml portion passed, the rest of the sample was introduced into the reservoir. 5 ml portions were used to rinse the cylinder used for collecting the reduced sample. After 20 ml was discarded, the following 15 ml was collected. To this 15 ml, 1 ml of the sulphanilamide solution was added, shaken, and left to stand for 2 minutes. 1 ml of the dihydrochloride was then added and the contents were shaken well. After 1 hour, the absorbance was determined using PYE Unicam SP6-250 visible spectrophotometer at a 543 nm, in 1 cm cuvette.

Capabilities:

The range of this method is between 0.05-60 µg at l^{-1}. The concentration levels and standard deviations for 10 samples at each level are:

40 ± 0.26 µg at NO_3^--Nl^{-1}

20 ± 0.12 µg at NO_3^--Nl^{-1}

1.0 ± 0.04 µg at NO_3^--Nl^{-1}

Establishment of the calibration curve

Nitrate standard was made up by dissolving 1.01 g KNO_3 in 1 litre of distilled water. 1 ml of this solution was made up to 1 litre by synthetic seawater. Various volumes of the latter were diluted with synthetic seawater to give different concentrations of nitrate ranging from 0 (synthetic seawater only) to 5 µg at NO_3-Nl^{-1}. These were treated and reduced as mentioned previously.

The corrected absorbances were plotted against their equivalent nitrate concentrations (Fig. 2.4).

A calibration factor (F) was determined and used for the direct calculation of nitrate in seawater.

Nitrite-nitrogen

The method used here was described by Strickland and Parsons (1972), based on the procedure described by Bendschneiber and Robinson (1952).

The nitrite in seawater was allowed to react with sulphanilamide in an acid solution. The resulting diazo compound was then reacted with N-(1-naphthyl)-ethylendiaminedihydrochloride solution to form a highly-coloured azo dye.

Reagents:

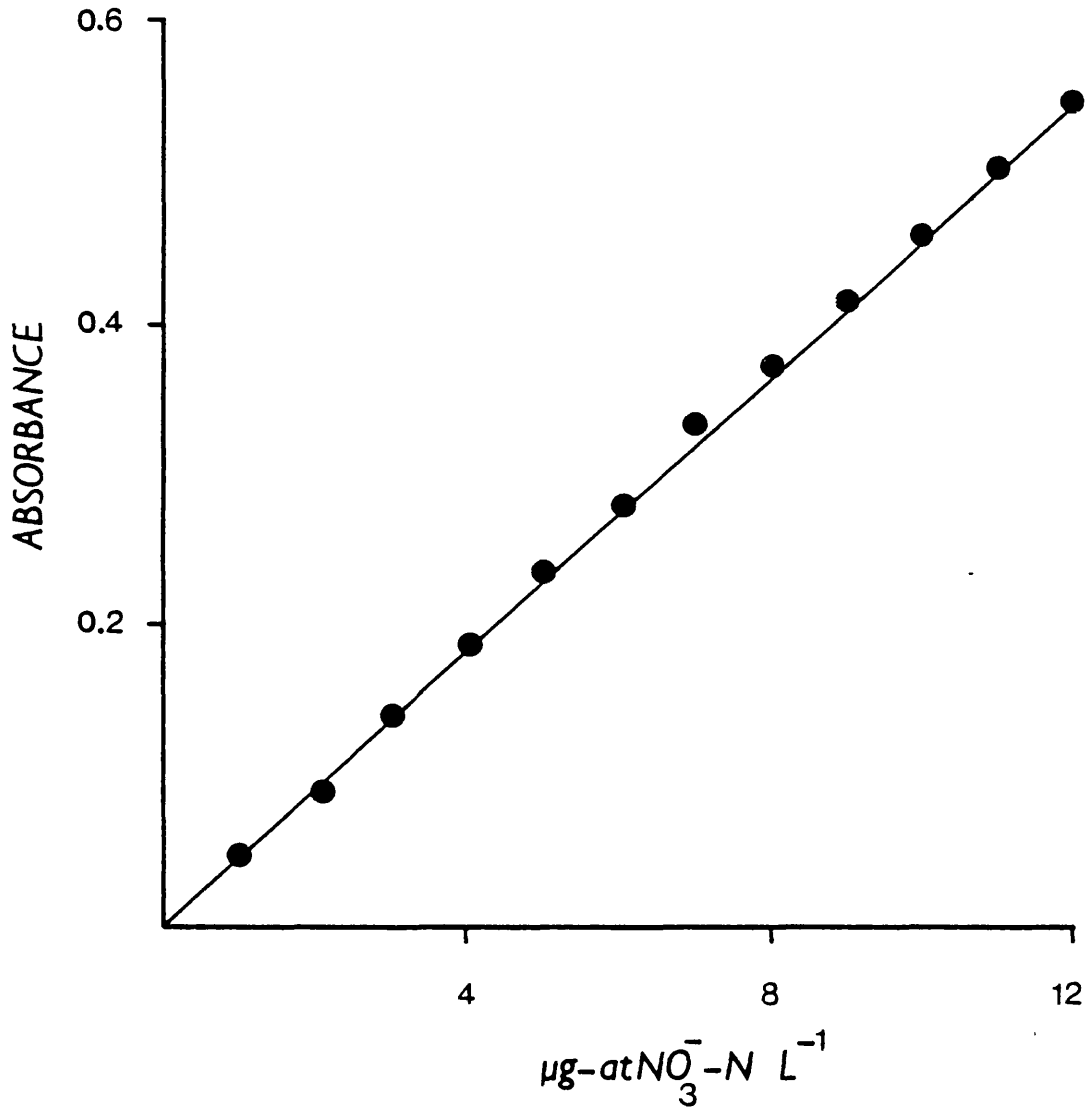
N-(1-naphthyl)-ethylendiaminedihydrochloride solution:

0.5 g of the dihydrochloride was dissolved in double distilled water and diluted to 500 ml.

Sulphanilamide solution:

5 g sulphanilamide dissolved in a solution of 50 ml of 12 N-HCl and 300 ml of double distilled water, then diluted to 500 ml with double

Figure 2.4 Nitrate calibration curve


0.6
0.4
0.2
ABSORBANCE
4
8
12
μg-at NO_3^--N L^{-1}

distilled water.

Procedure:

50 ml sample of the filtered seawater was placed in 50 ml graduated cylinder. To this sample 1 ml sulphanilamide was added. The contents were shaken well and then kept for 2 minutes to react. 1 ml of the dihydrochloride solution was then added and the contents mixed together. This process was repeated for all the samples and for the reagent blank.

After 1 hour, the absorbance was read in a spectrophotometer, against distilled water as a blank in a 1 cm cell at 543 nm.

Capabilities:

Range: 0.01-2.5 µg at l^{-1}

Precision at the 1 µg at l^{-1} level

The correct value lies in the range, mean of

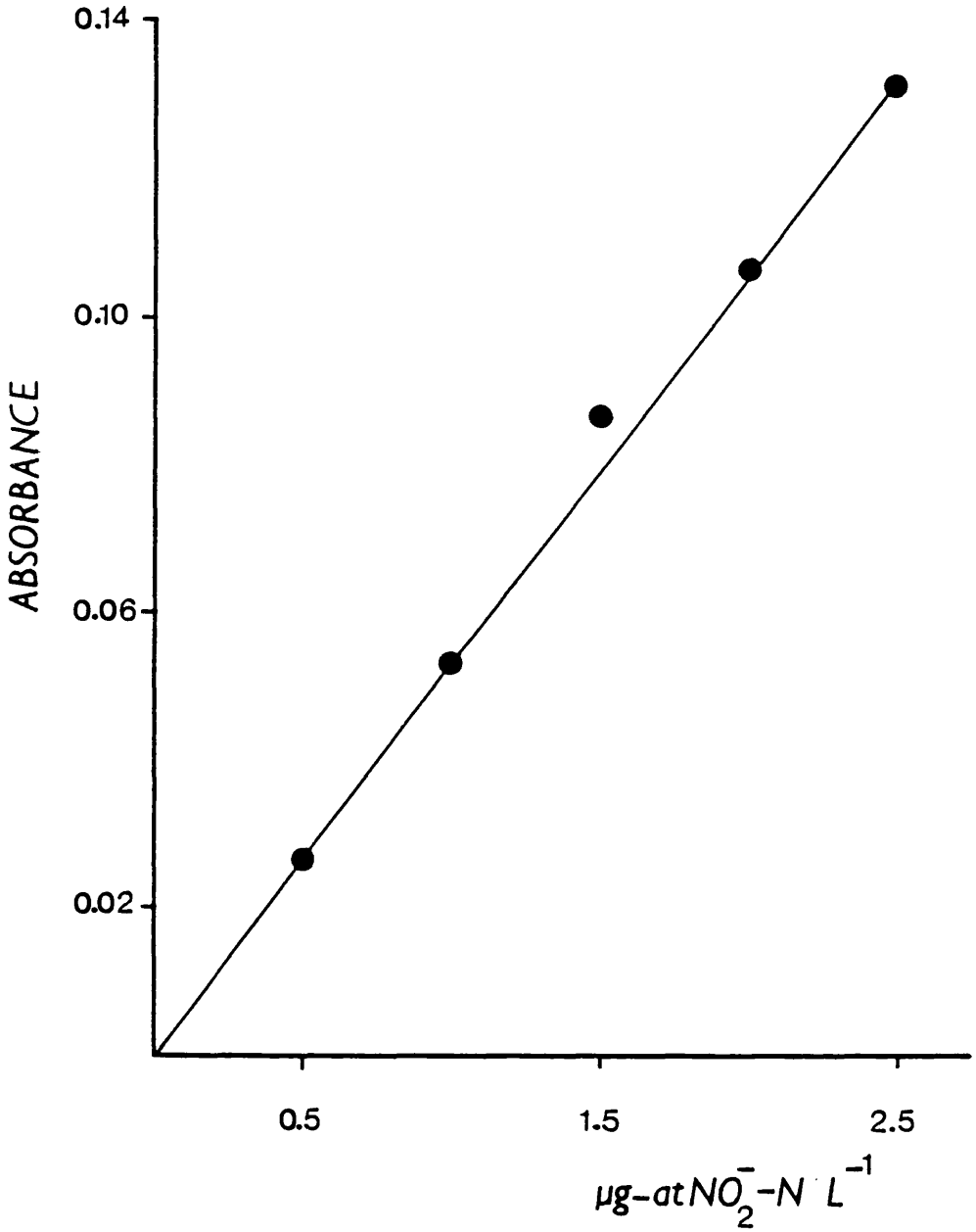
$\underline{n}$ determinations $\pm$ $0.03/n^{\frac{1}{2}}$ µg at l^{-1}

Establishment of the calibration curve

Anhydrous sodium nitrite was dried at 100°C for 1 hour. 0.345 g was dissolved in 1 litre of double distilled water. This gave a concentration of 5 µg at N ml^{-1}.

Various volumes of this standard diluted with double-distilled water to 50 ml portions with concentrations between 0 (double-distilled water only) and 0.5 µg at NO_2-N.l^{-1}. The procedure described above was followed and the corrected extinctions obtained were plotted against their equivalent concentrations (Fig. 2.5). A calibration factor (F) was obtained from the curve as follows:

Figure 2.5 Nitrite calibration curve


0.14
0.10
0.06
0.02
ABSORBANCE
0.5
1.5
2.5
μg-at NO_2^--N L^{-1}

$$F = \frac{20}{E_s - E_b}$$

E_s = extinction of sample, E_b = extinction of blank

Ammonia-nitrogen

The method used in this study was described by Koroleff (1976). This method is based on the reaction of ammonia in moderately alkaline solution with hypochlorite to give monochloramine. In the presence of phenol, nitroprusside ions and excess hypochlorite, the monochloramine gives indophenol blue.

Reagents:

Sodium hydroxide (0.5N):

20 g of sodium hydroxide dissolved and made up to 1 litre with deionized water.

Magnesium sulphate solution:

50 g of magnesium sulphate heptahydrate was dissolved in about 100 ml of deionized water. 0.5 N sodium hydroxide was added until a slight precipitate was formed. With the aid of anti-bumping granules, the solution was boiled to evaporate any traces of ammonia. When the volume was less than 100 ml, it was cooled and made up to 100 ml with deionized water.

Phenol reagent:

38 g of phenol and 400 mg of disodium nitroprusside dihydrate were dissolved, then diluted to 1 litre with deionized water.

Sodium thiosulphate solution:

24.82 g of $Na_2S_2O_3.5H_2O$ was dissolved in 1000 ml double-distilled water to give 0.1N solution.

Sodium hypochlorite solution:

The available chlorine in the sodium hypochlorite stock solution was determined as follows:

0.5 g potassium iodide was dissolved in 50 ml of 1N sulphuric acid. 1.0 ml of the hypochlorite stock was added and the iodine librated was titrated against 0.1N thiosulphate using starch as indicator.

1 ml 0.1N thiosulphate = 3.54 mg active chlorine

Using the above equation, hypochlorite reagent containing 150 mg available chlorine per 100 ml was prepared using 0.5 N NaOH.

Tri-sodium citrate solution:

240 g of tri-sodium citrate dihydrate was dissolved in 500 ml double-distilled water. This solution was then made alkaline by the addition of 20 ml of 0.5 N NaOH. Ammonia was removed by boiling. The solution was made up to 500 ml with deionized water.

Procedure:

In a 50 ml volumetric flask, 35 ml of the sample was poured. To this sample, 1 ml of the citrate solution, 1 ml of the phenol reagent and 1 ml of the hypochlorite reagent were added. The contents were mixed well by swirling between additions. The sample was kept in the dark overnight. The extinctions of the sample and blank were measured in 1 cm cuvette against acidified distilled water at 630 nm.

Capabilities:

Range: 0.01-10 µg at NH_4^+-Nl^{-1}

Precision of the method at 1 µg at l^{-1} level

The correct value lies in the range of mean of

$\underline{n}$ determinations $\pm$ $0.1/n^{\frac{1}{2}}$ µg at l^{-1}

Calibration

53.5 mg NH_4Cl was diluted to 100 ml with deionized water. This stock was diluted again with deionized water to give a concentration of 1 µg at Nl^{-1}. Three 35 ml portions of this new solution in addition to two 35 ml portions of deionized water were treated as in the procedure outlined above.

The mean of the reagent blank extinctions was substracted from the mean of the samples extinctions.

The result (A) was applied in the following equation:

$$F = \frac{1.0}{A}$$

where F was the calibration factor used in the determination of ammonia-nitrogen in seawater.

Silicate silicon

The method used for silicate analysis was the one described by Strickland and Parsons (1972).

Using this method, the seawater sample is reacted with molybdate in order to form silicomolybdate, phosphomolybdate and arsenomolybdate complexes. The silicomolybdate complex is then reduced by a reducing solution to give blue colour.

Reagents:

Molydbdate reagent:

4.0 g of ammonium paramolybdate was dissolved in 300 ml of double-distilled water, and then 12 ml of concentrated hydrochloric acid was added. This mixture was made up to 500 ml with double distilled water.

Metol-sulphite solution:

6.0 g of anhydrous sodium sulphite was dissolved in 500 ml of double-distilled water. To this solution, 10 g of metol was then dissolved. This solution was filtered through a No. 1 Whatman filter paper.

Oxalic acid solution:

Saturated oxalic acid was prepared by dissolving 50 g of oxalic acid dihydrate in 500 ml double-distilled water.

Sulphuric acid solution (50% v/v):

250 ml of concentrated sulphuric acid was added slowly to 250 ml of double distilled water, kept cool by running water.

Reducing reagent:

100 ml of metol-sulphite solution was mixed with 60 ml of oxalic acid. 60 ml of 50% sulphuric acid was added slowly with mixing. The mixture was made up to 300 ml with double distilled water.

Procedure

10 ml of molybdate solution was added to a dry 50 ml volumetric plastic flask, fitted with a stopper. 25 ml of seawater sample at room temperature was added to the 10 ml molybdate reagent in the flask. The

mixture was allowed to stand for 15 minutes. 15 ml of the reducing reagent was added to the mixture to make the volume up to 50 ml. The solution was then allowed to stand for 2 hours to complete the reduction. The extinction of the reagent blank and the sample were measured against double-distilled water in 1 cm cuvette at 810 nm.

Capabilities:

Range: 0.1 to 140 µg at l^{-1}.

Precision at the 10 µg at l^{-1} level:

The correct values lie in the range:

mean of $\underline{n}$ determinations $\pm$ $0.25/n^{\frac{1}{2}}$ µg at l^{-1}.

Establishment of the calibration curve

Standard silicate solution was made up by dissolving 0.96 g silicofluoride in 100 ml double-distilled water. It was then diluted to 1 litre with double-distilled water.

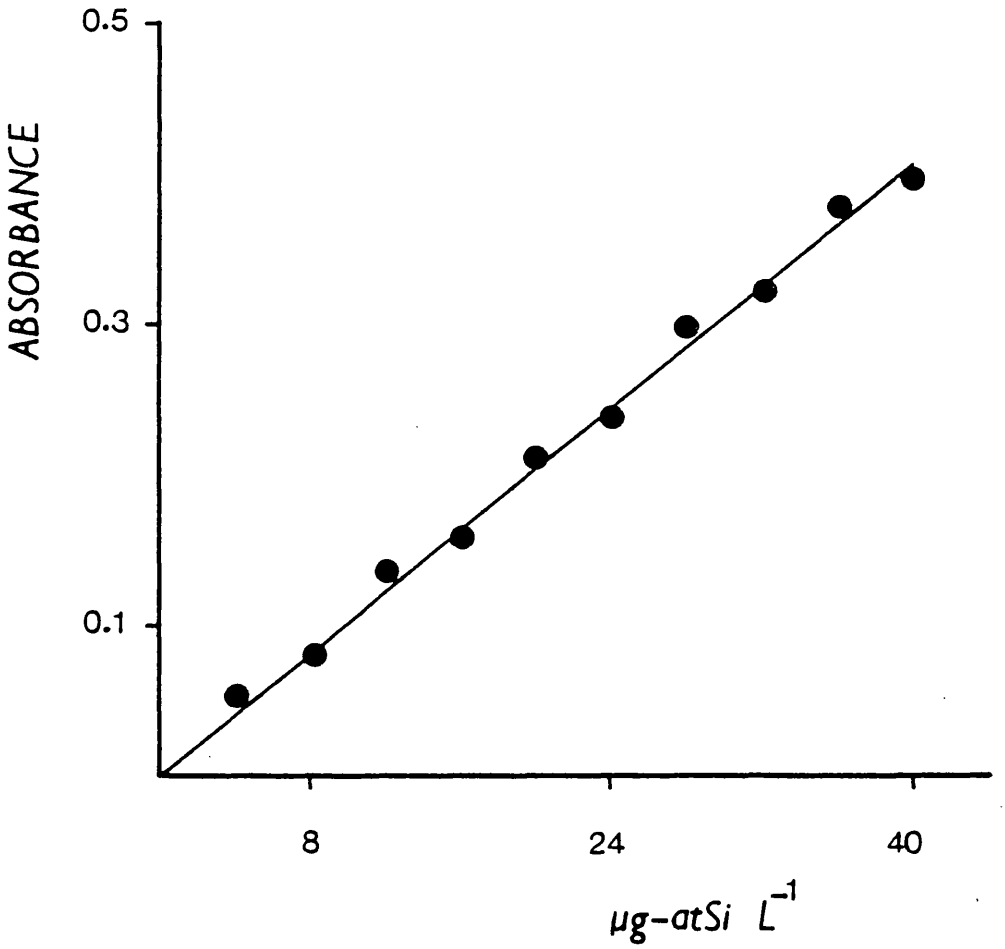
1 ml ≙ 5 µg-at Si

Various volumes of the above solution were made up to 25 ml portions with synthetic seawater to give 0-96 µg at Si l^{-1}.

These standards were treated as described in the procedure and the resulting extinctions were plotted against their equivalent concentrations (Fig. 2.6). A calibration factor (F) was calculated from the curve using the formula:

$$F = \frac{100}{Es-Eb}$$

Figure 2.6 Silicate calibration curve


0.5
0.3
0.1
ABSORBANCE
8
24
40
μg-atSi L⁻¹

Phosphate-phosphorus

The method used here was described by Strickland and Parsons (1972) and was taken from the original work by Murphy and Riley (1962).

The theory of the method is to allow the seawater to react with molybdic acid, ascorbic acid and trivalent antimony. The resulting complex is reduced to give a blue colour.

Reagents:

Ammonium molybdate solution:

15 g of ammonium paramolybdate was dissolved in 500 ml of double-distilled water.

Sulphuric acid solution:

140 ml of concentrated sulphuric acid was added to 900 ml of double-distilled water.

Ascorbic acid solution:

27 g of ascorbic acid was dissolved in 500 ml of double-distilled water.

Potassium antimonyl-tartrate solution:

0.34 g of potassium antimonyl-tartrate was dissolved in 250 ml of double distilled water.

Mixed reagent:

100 ml of the ammonium molybdate was mixed with 250 ml sulphuric acid, 100 ml ascorbic acid, and 50 ml of potassium antimonyl-tartrate solutions. This mixed solution was prepared immediately before use.

Procedure

Into 100 ml glass bottle, 50 ml seawater at room temperature was introduced. 10 ml of the freshly prepared mixed reagent was added. After 30 minutes the extinctions of reagent blank and the sample were measured in 1 cm cuvette against distilled water at 885 nm.

Capabilities:

A range of phosphate concentrations between 0.03-5 µg at l^{-1} can be determined.

Precision at the 3 µg at l^{-1} level:

The correct value lies in the range:

mean of n determinations $\pm$ $0.03/n^{\frac{1}{2}}$ µg at l^{-1}.

Establishment of the calibration curve

A phosphate standard solution was prepared by dissolving 1.361 g of anhydrous potassium dihydrogen phosphate in 1 litre of double-distilled water.

1 ml ≡ 10 µg at P

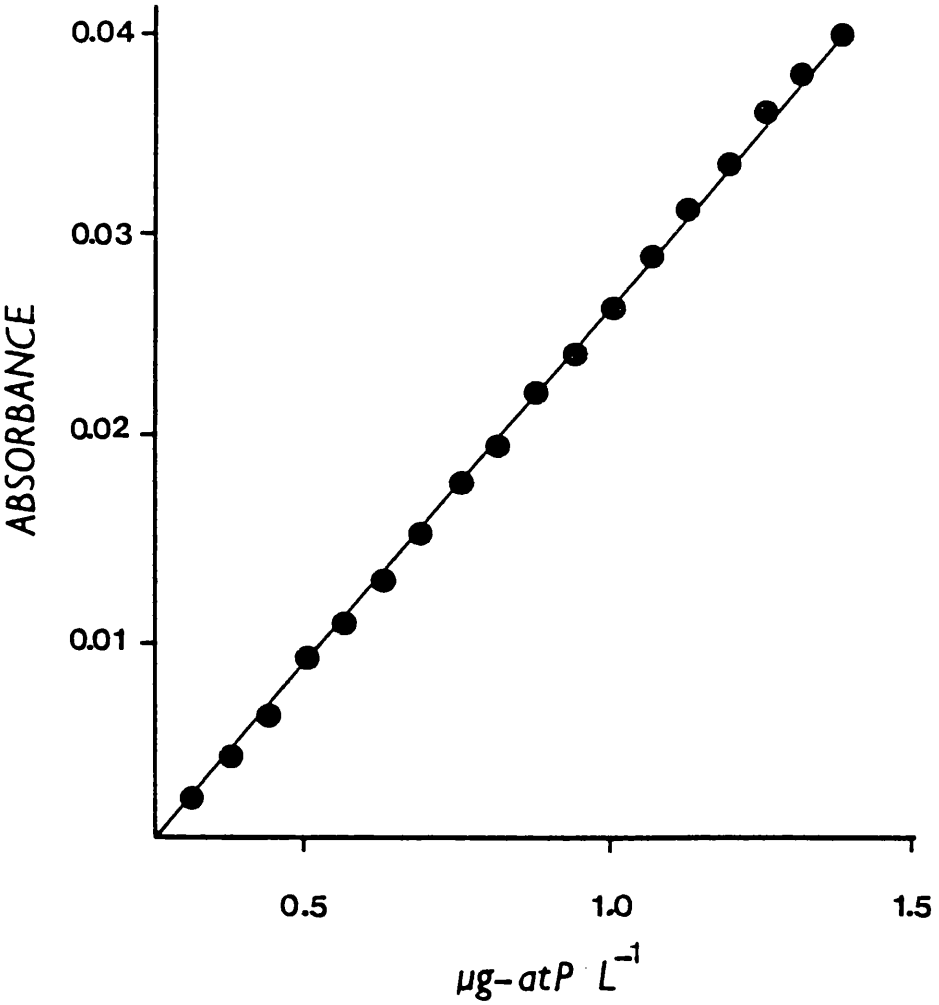
Various volumes of this standard were diluted to 50 ml with double-distilled water to give a range of concentrations between 0-14 µg at P l^{-1}.

The steps described in the procedure were followed and the corrected extinctions were plotted against their equivalent concentrations (Fig. 2.7).

A calibration factor (F) was calculated from the calibration curve by the following equation:

$$F = \frac{3.00}{Es-Eb}$$

Figure 2.7 Phosphate calibration curve


0.04
0.03
0.02
0.01
ABSORBANCE
0.5
1.0
1.5
μg-atP L⁻¹

CHAPTER III

ENVIRONMENTAL FACTORS

INTRODUCTION

Abiotic factors in the marine environment have been known for a long time to affect phytoplankton production. By comparing the seasonal productivity cycles in the coastal waters and nutrient concentrations, it was observed that phytoplankton biomass and primary productivity diminish as nutrient concentrations were reduced to low levels in the late spring and early summer (Brandt, 1898; Marshall and Orr, 1928; Lillick, 1937).

This effect has not been restricted to the nutrients. Light, temperature, salinity and water column stability could limit the phytoplankton production as well as nutrients (Braarud and Klem, 1931; Marshall and Orr, 1948).

Abdullah et al. (1973) made a detailed study in the Bristol Channel describing the circulation and hydrography in the channel and their role in the distribution of the nutrients affecting phytoplankton growth. They suggested two distinct regions separated by a line drawn from Mumbles Head to Foreland Point. The nutrient distribution and salinity in the eastern region were mainly controlled by river runoff. High levels of nutrients were found in this region. On the other hand, the nutrient level in the western region was found to be low. This was suggested to be due to the mixing between the river runoff and the Celtic Sea water. Abdullah et al. (1973) suggested that the main source of nutrients in the Bristol Channel was river runoff.

The cycling and variation of physical parameters, nutrients and plankton in Swansea Bay have been studied by Pearce (1967), Abdullah et al. (1973), Gabriel (1973), Isaac (1974) and Paulraj (1974).

The plankton and water chemistry were studied intensively by

Paulraj (1974) but the study was restricted to the area close to Mumbles pier.

More studies have been carried out recently by Morris and Mantoura (1980), Joint (1980), Humphrey et al. (1980). Although these studies were intensive, they were mainly carried out during selected times of the year and in the outer borders of Swansea Bay.

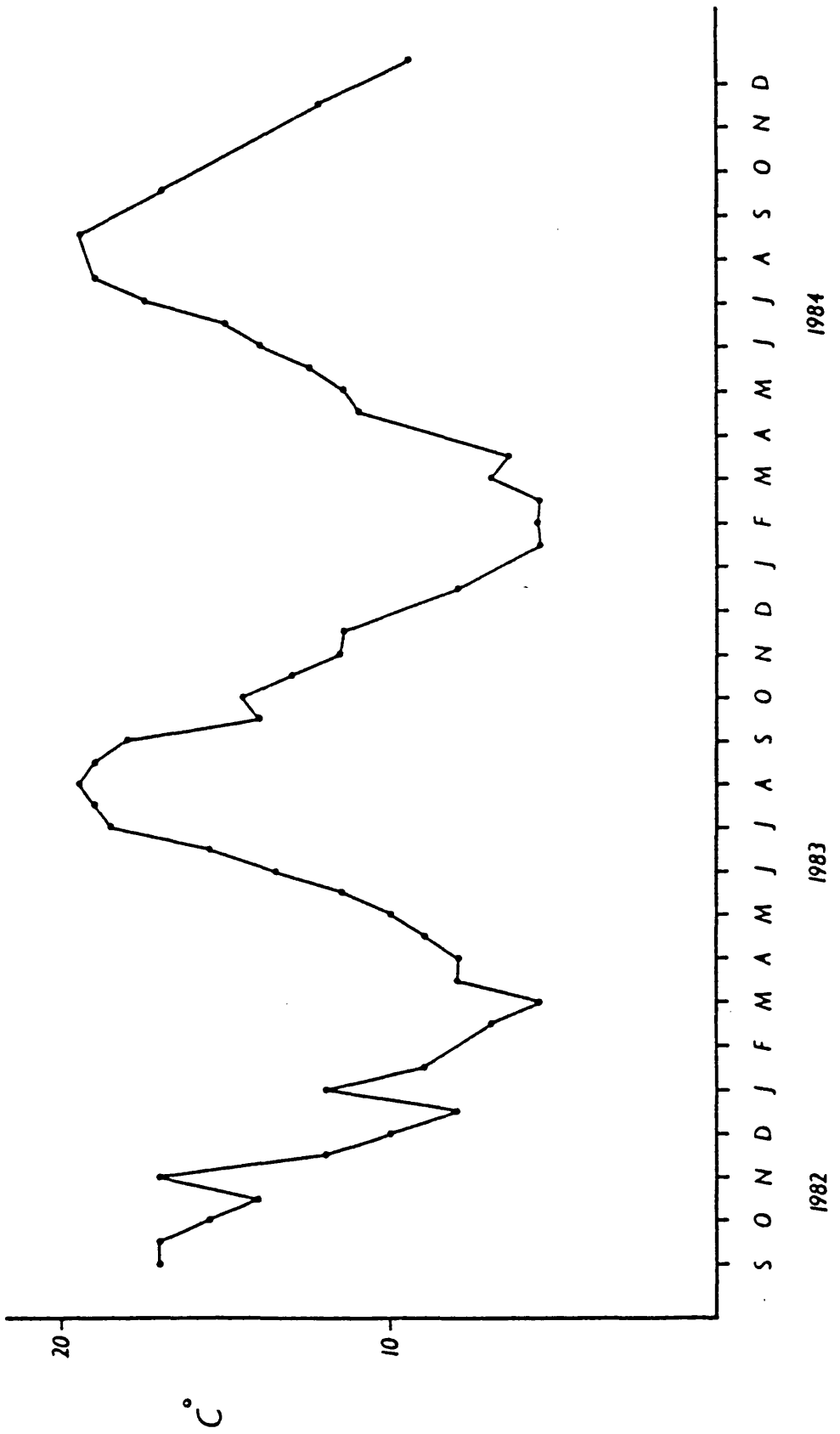
The present study was conducted for more than two annual cycles in the inshore waters of Swansea Bay. The reason for the study of the chemical and physical factors in the present study was to provide a basis of parameters which could affect the primary production of phytoplankton in Swansea Bay.

RESULTS

1. Seawater Temperature

The variation in water temperature ranged from 5.5°C to 19.5°C (Fig. 3.1). From 17°C in the autumn (1982) the temperature declined through the winter (1983) down to 5.5°C in March. From that time, the temperature increased steadily throughout the spring and summer reaching its maximum in August (19.5°C). After it reached its highest value in August, the temperature declined steadily throughout the autumn and winter until it reached a minimum of 5.5°C in January-February (1984). Once again the water temperature started to increase at the beginning of the spring until it reached its annual maximum in August (19.5°C). After August it began to decrease until it reached 9.5°C in December when the sampling was stopped.

Figure 3.1 Seawater temperature


20
10
°C
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

2. Air Temperature

The maximum and minimum air temperature during the period of sampling were supplied by Penmaen Weather Station (Fig. 3.2). These air temperatures fluctuated in a similar pattern to that of surface water temperature.

a. Maximum Air Temperature

At the time when the sampling programme was first started the temperature was 18.9°C (September, 1982). During the following months, the temperature decreaseed until it reached the minimum of 5.6°C (February, 1983). Thereafter, the temperature started to increase until it reached the annual maximum of 25.9°C (July, 1983). This cycle was repeated again with minimum temperature of 5.8°C (January, 1984) and maximum temperature of 21.3°C (July, 1984).

b. Minimum Temperature

The minimum temperature throughout the years of sampling ranged from 0°C to 19.3°C.

The temperature of September, 1982 was 13°C. This temperature dropped steadily down to 0°C in December. From January, 1983 the temperature started to increase until it reached the highest minimum temperature ever encountered during this study of 19.3°C in July. The decrease of temperature afterwards started a new but similar annual cycle.

3. Monthly Sunshine Hours

The variation in sunshine hours (Fig. 3.3) was characterized by alternative periods of low and high monthly sunshine hours. The period from

Figure 3.2 Maximum and mimimum air temperature

● Maximum

• Minimum

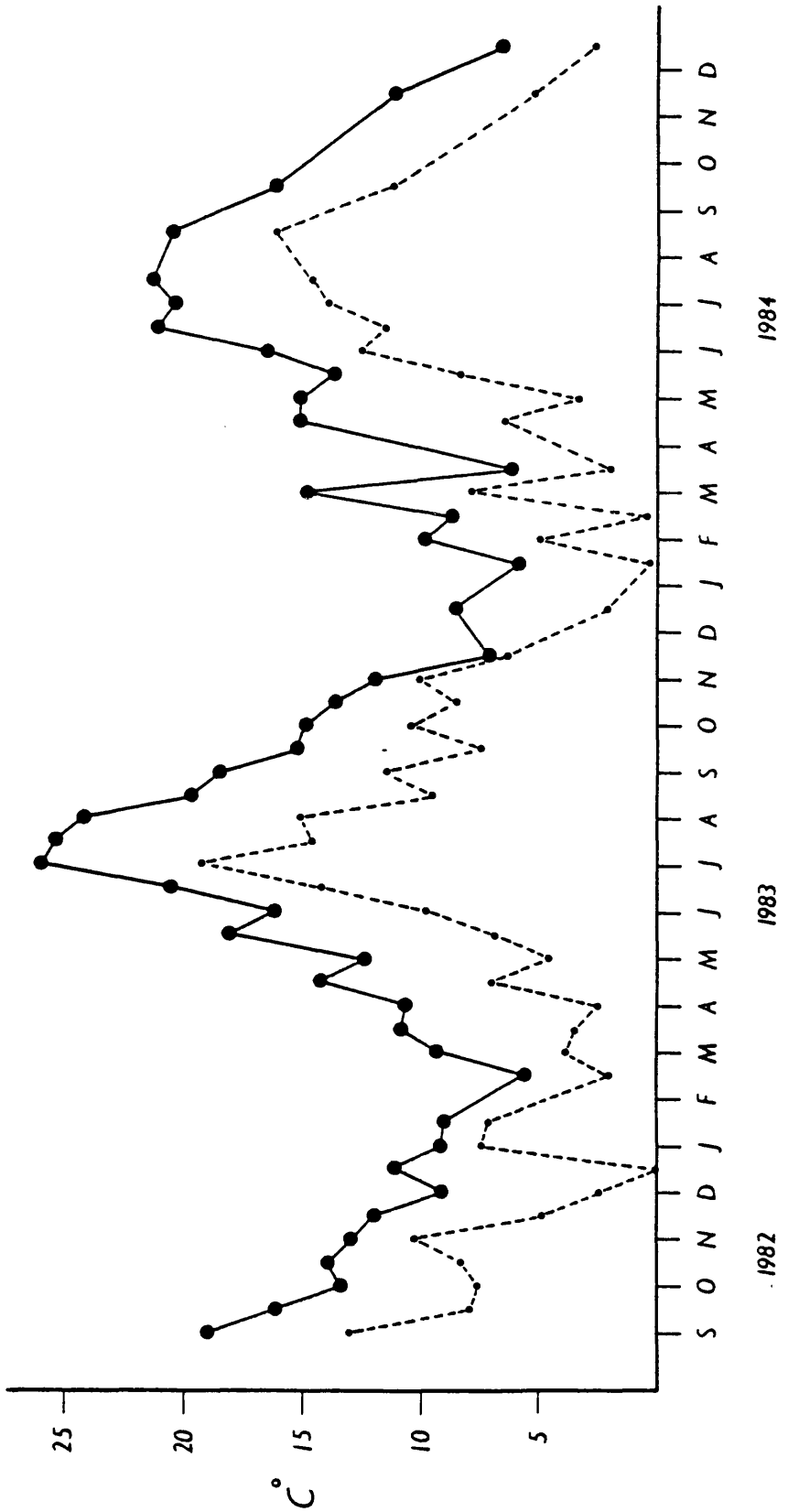
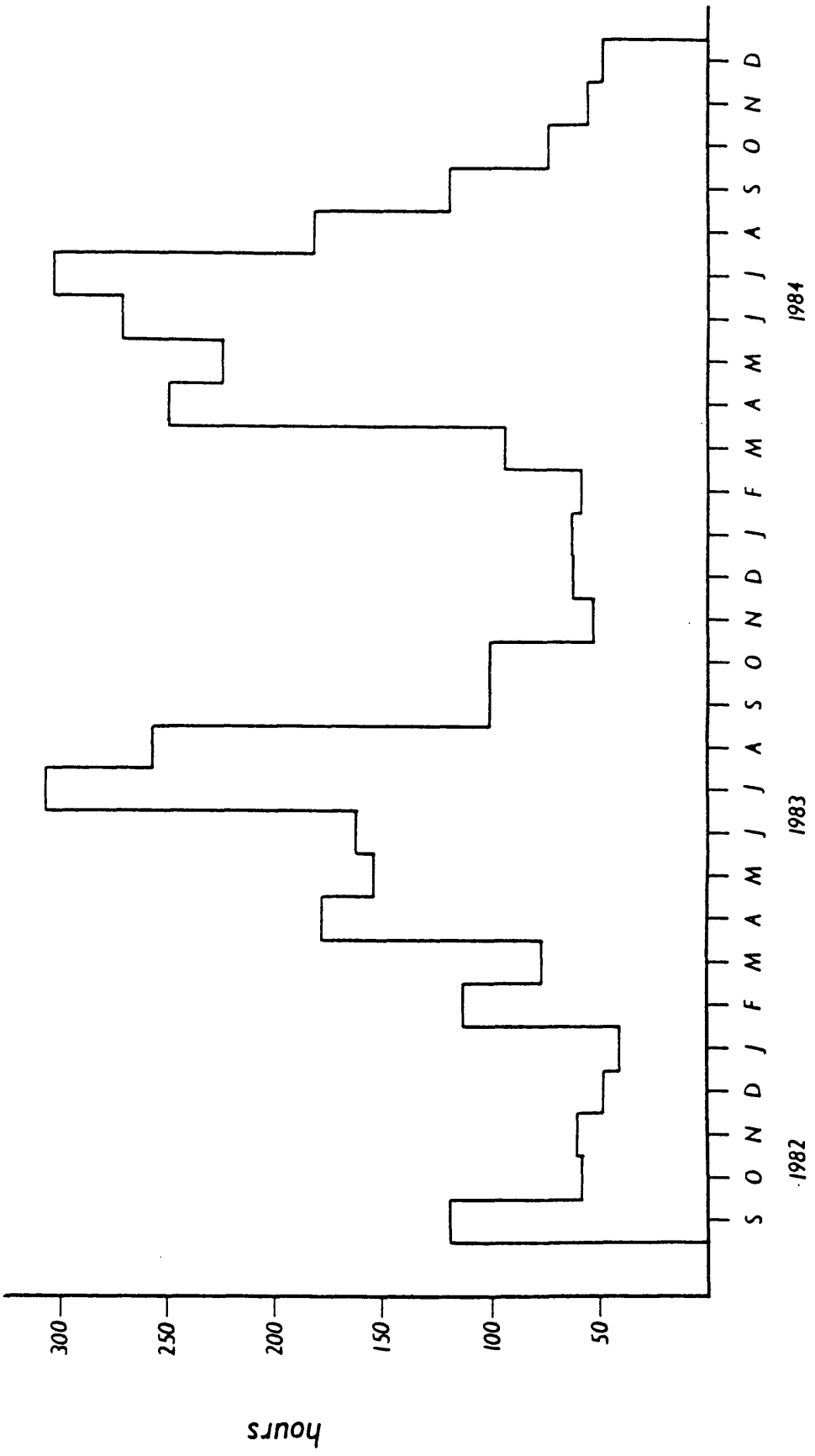

25
20
15
10
5
°C
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

Figure 3.3 Total monthly sunshine hours


hours
300
250
200
150
100
50
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

September, 1982 to January, 1983 was characterized by a low number of sunshine hours. The total sunshine hours for that period was 382 hours with a daily average of 2.18 hours. The minimum value of sunshine hours was in January (41.2 hours). The period from February to October, 1983 had high values of monthly sunshine hours. The total sunshine hours of that period was 1450.3 hours with a daily average of 5.37 hours.

July was the month with the highest number of sunshine hours (307.6) with a daily average of 10.25 hours. This period was followed by one with a low number of sunshine hours (November, 1983 to February, 1984). The total for that period was 237.3 hours with a daily average of 1.97 hours. The period of high numbers of sunshine hours extended from March to September, 1984. The total sunshine hours for this period was 1437.8 hours and the daily average was 6.84 hours. Thereafter, the daily sunshine hours began to decrease.

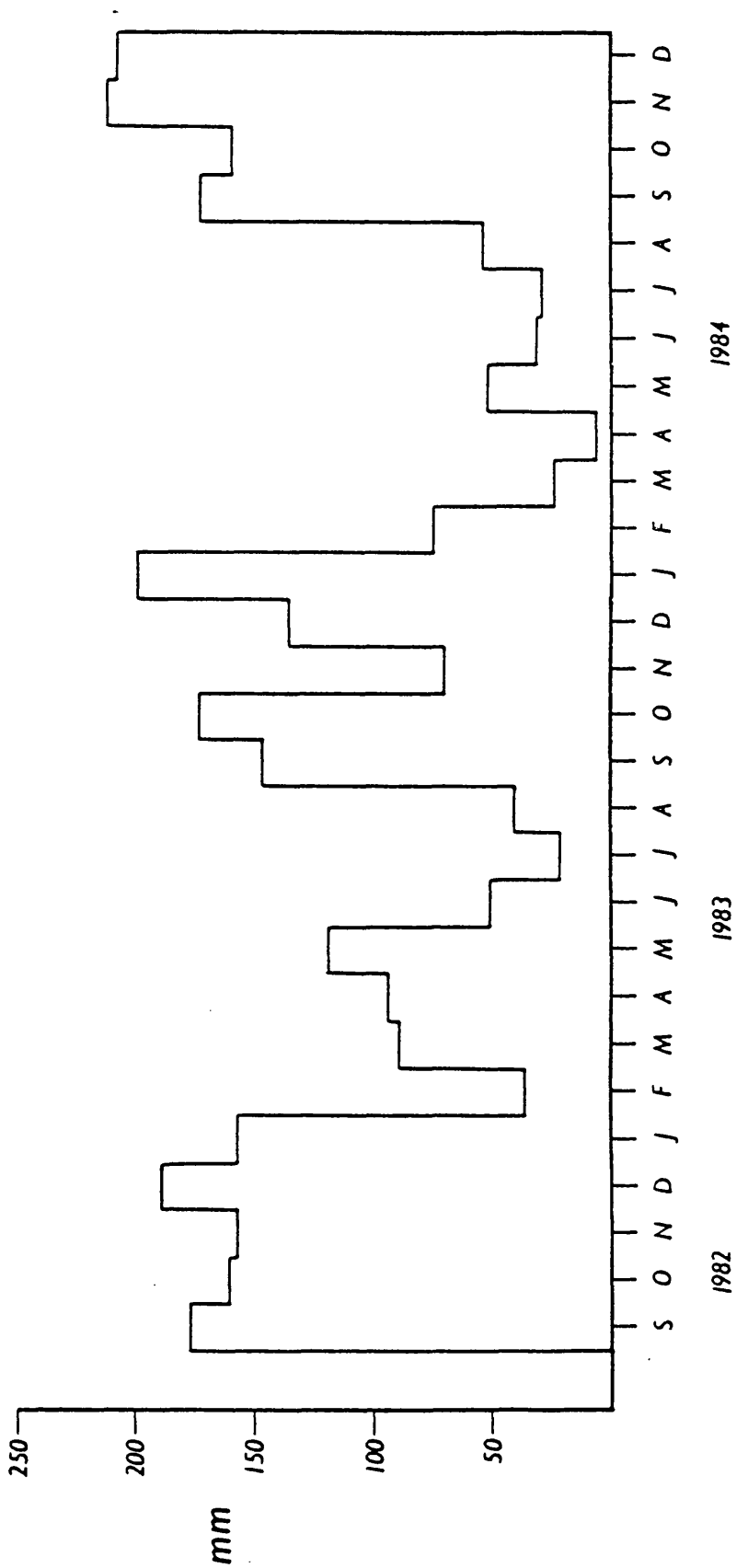
4. Monthly Rainfall

During this study, three periods with high monthly rainfall have been found (Fig. 3.4). These periods occurred mainly during the autumn and winter months.

The first period, from September, 1982 to January, 1983, had a monthly average of 168 mm. The period which extended from February to August, 1983 recorded a monthly average of 67.15 mm. The next rainy season started in September, 1983 continuing until January, 1984 with a monthly average of 144.94 mm. This period was followed by one with low monthly average (38 mm), extending from February to August, 1984.

Once again the summer period, with low monthly averages of rain, was followed by a period extending from September to December, 1984

Figure 3.4 Total monthly rainfall


250
200
150
100
50
mm
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

with a monthly average of 187.7 mm.

5. Salinity

i. Station A

In this station, salinity was variable over a range of 25.9-28.6‰ between September, 1982 and May, 1983 (Fig. 3.5). Two exceptions were observed during that period; low value in November, 1982 (23.6‰), and another low value in January, 1983 (24.7‰).

The salinity started to increase steadily from May, 1983 until it reached a value of 30‰ in July, 1983.

A sudden drop in salinity was observed during the period from August-September, 1983. The minimum value in this period was 25.5‰ (August). After that period the salinity returned to its normal level of ca. 28-29‰. There was a sudden drop in the value of salinity from the normal level down to a very low value of 23.4‰ in February, 1984. Thereafter, the salinity returned to its normal level until November, 1984 when it dropped again.

ii. Station B

Salinity was mainly observed over a range of 24-31.2‰ (Fig. 3.6). The salinity value of 28.5‰ in September, 1982 started to decrease steadily until it reached an exceptional value of 16.3‰ in November, 1982. This was followed in January, 1983 by a relatively low salinity value of 24‰. From January to April, 1983 the salinity remained around 28‰. As in Station A, the period from August to September, 1984 was characterized by low salinity values (25.5‰ in August). From October, 1983 the salinity increased again until it reached ca. 29.5‰. It remained at this level until February, 1984 when there was a sudden

Figure 3.5 Salinity (Station A)

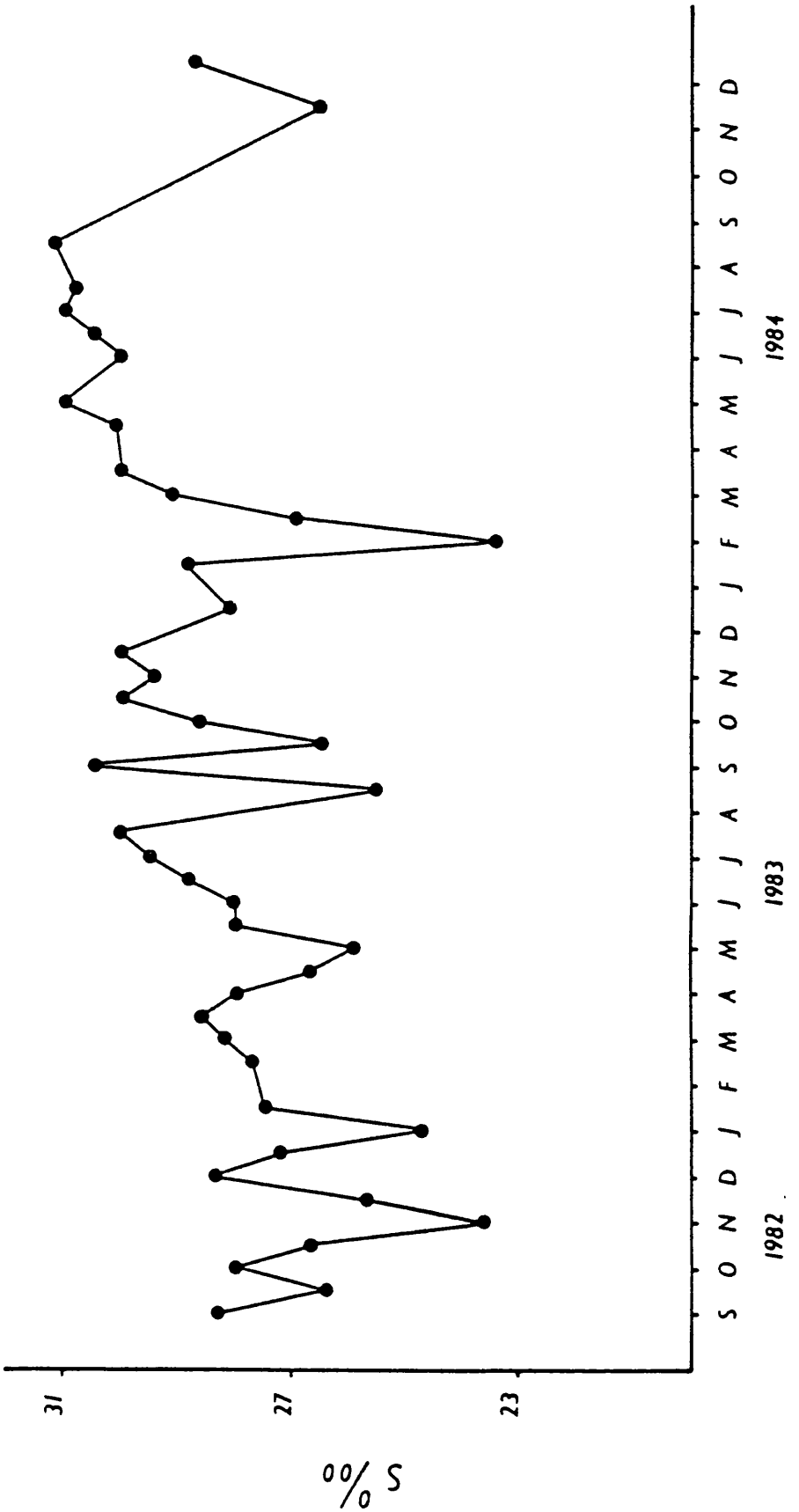
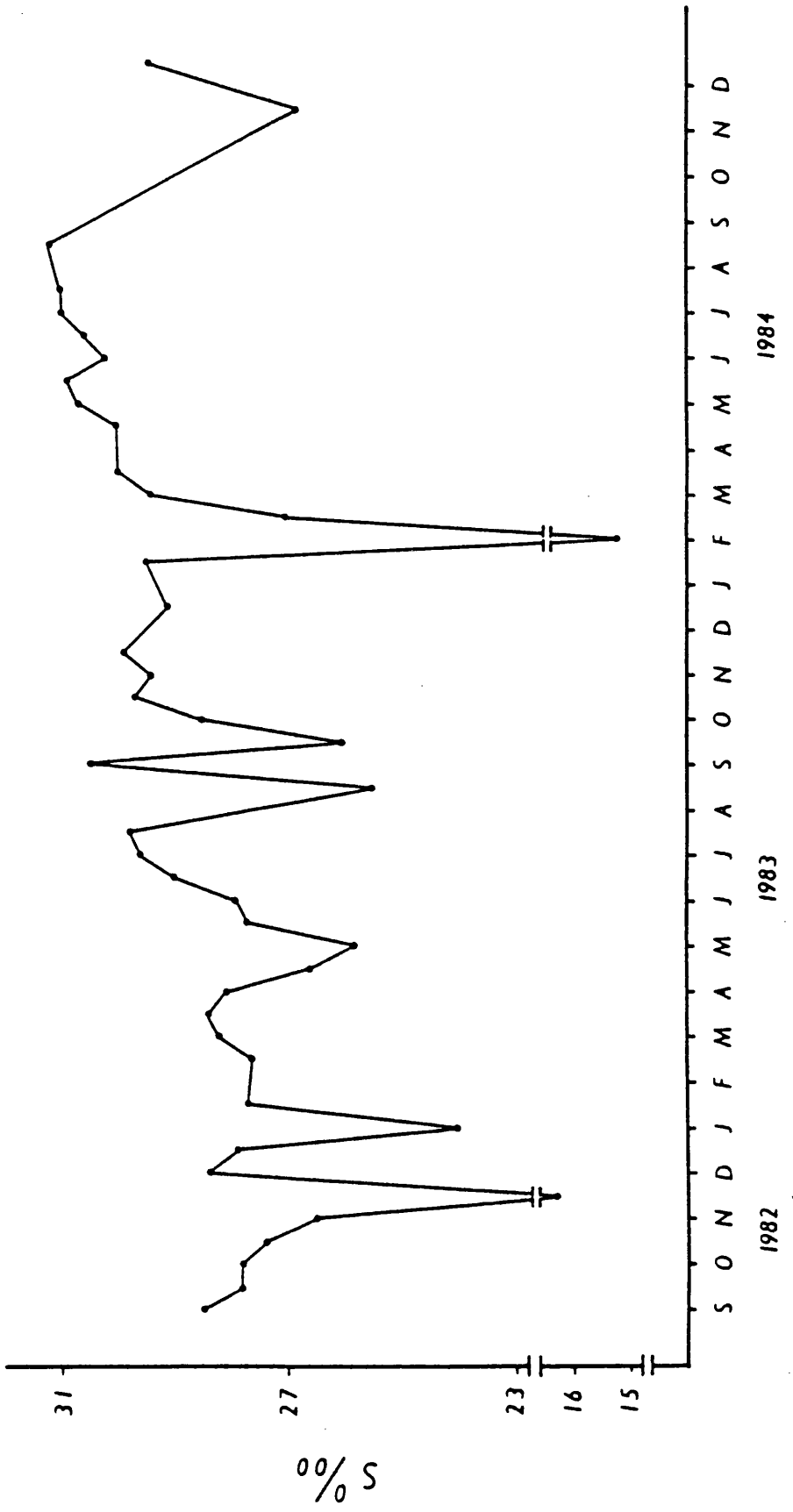

‰ S
31
27
23
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

Figure 3.6 Salinity (Station B)


31
27
23
16
15
‰ S
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

drop in salinity down to an exceptional value of 15.2%o. This drop was followed by a steady increase during the spring and summer months until the salinity was around 30%o. The salinity remained at this level until November, 1984 when there was a drop down to 26.8%o.

6. Nitrate-nitrogen

The concentrations of nitrate-nitrogen in both stations were very similar during this study and there was no significant difference at any point (Fig. 3.7 and Fig. 3.8). For this reason their results will be described as one.

In September, 1982 nitrate-nitrogen concentration was 1.5 µg at l^{-1}. The concentration increased steadily until it reached a maximum of 49 µg at l^{-1} in November, 1982. Between November, 1982 and June, 1983, the concentration values remained around 34 µg at l^{-1}. By the end June, 1983 there was a sudden drop from ca. 36 µg at l^{-1} down to ca 6.7 µg at l^{-1}. This decrease in the concentration continued until it reached a minimum value of ca. 0.3 µg at l^{-1}. Thereafter nitrate concentration started to increase steadily until it reached the highest maximum in the study of ca. 66 µg at l^{-1}. This was followed by a steady decrease down to a minimum of 0.2 µg at l^{-1} (Station A), and 0.144 µg at l^{-1} (Station B) in May. These low values were followed by a steady increase until December, 1984 when it reached ca. 35 µg at l^{-1}.

7. Nitrite-nitrogen

i. Station A

Nitrite-nitrogen was observed over a range of ca. 0.019 µg at l^{-1} to 3.24 µg at l^{-1} (Fig. 3.9).

A September concentration of ca. 0.45 µg at l^{-1} was followed by a

Figure 3.7 Nitrate-nitrogen (Station A)

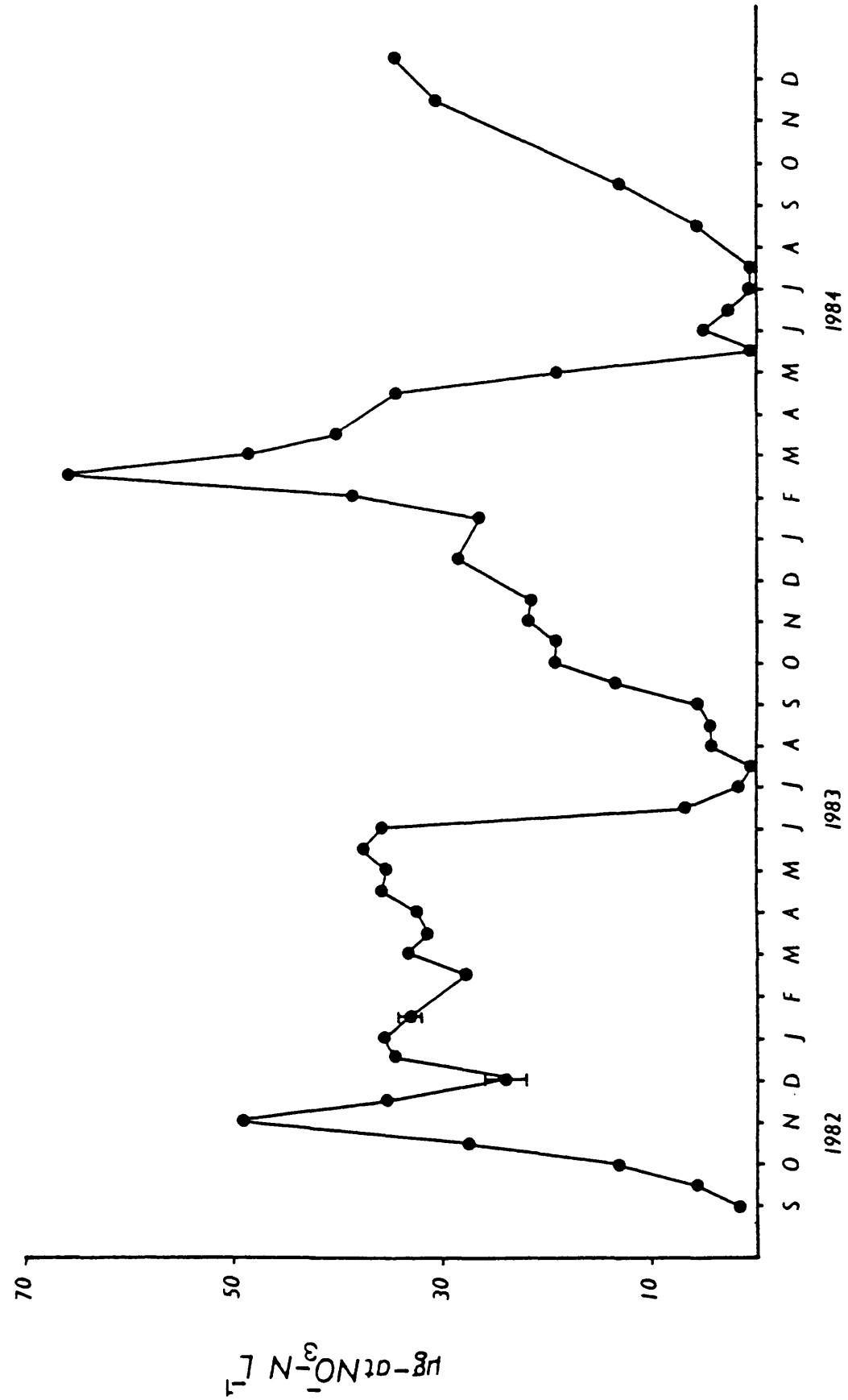

µg-at NO_3^--N L^{-1}
70
50
30
10
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

Figure 3.8 Nitrate-nitrogen (Station B)

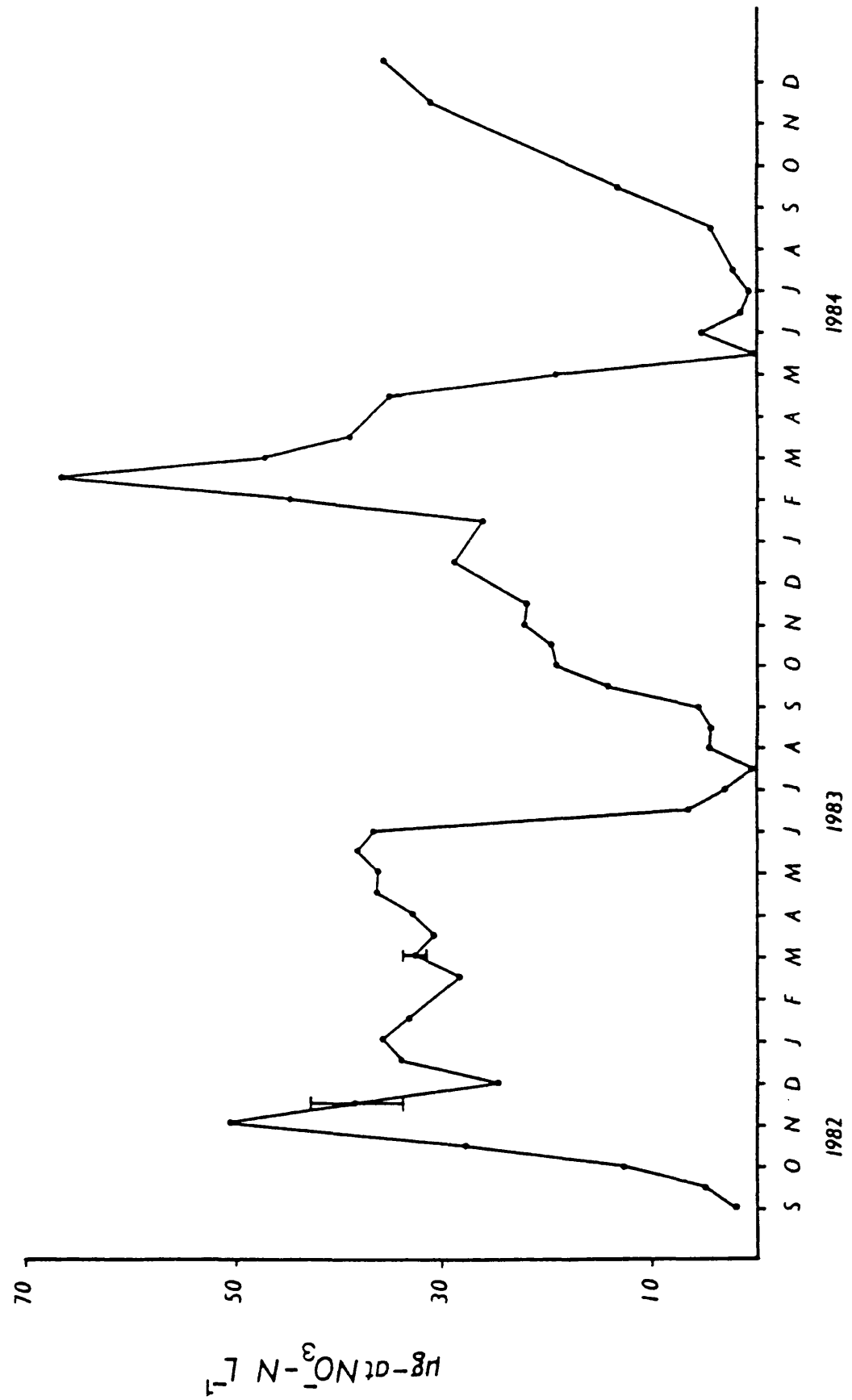
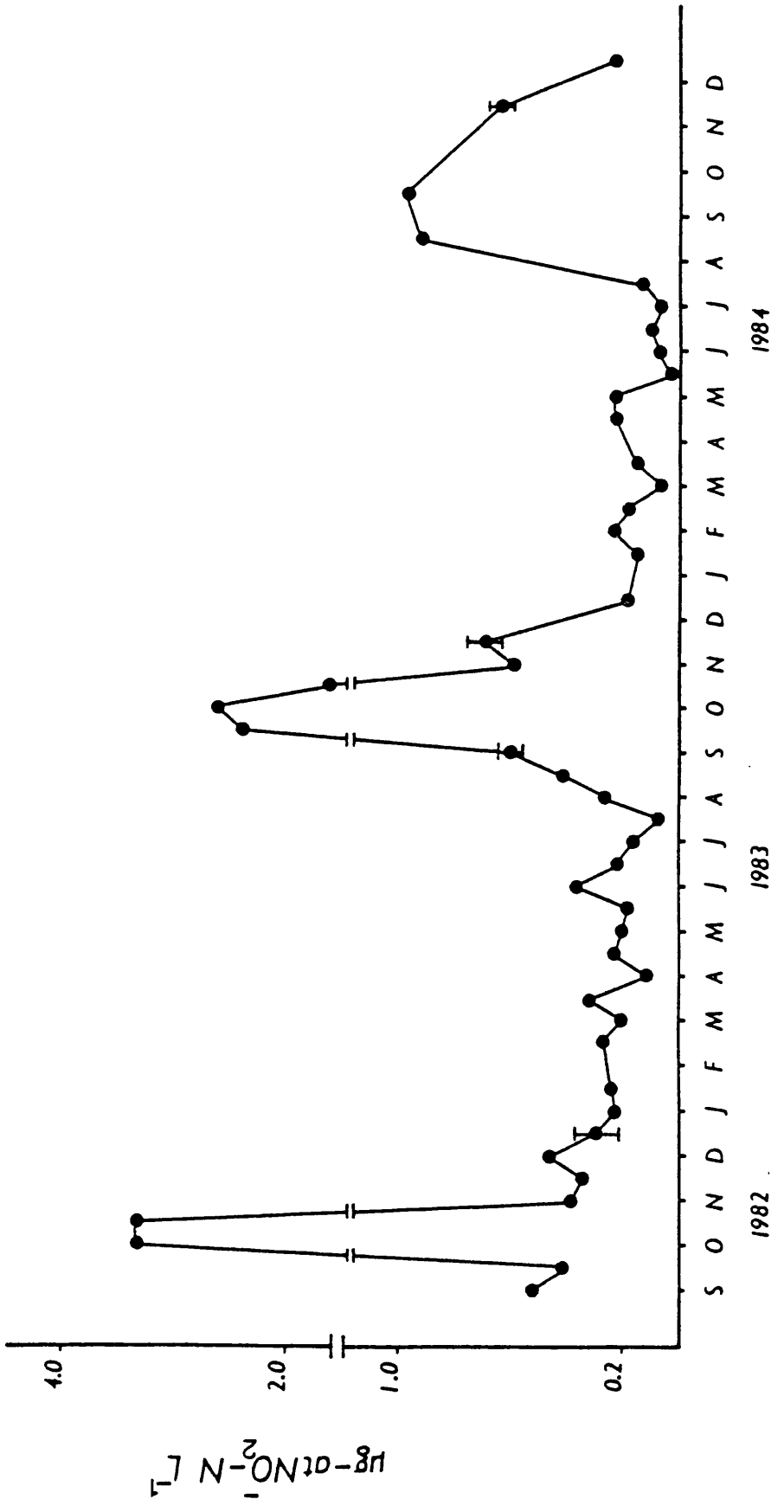

µg-at NO_3^--N L^{-1}
70
50
30
10
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

Figure 3.9 Nitrite-nitrogen (Station A)


μg-at NO_2^--N L^{-1}
4.0
2.0
1.0
0.2
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

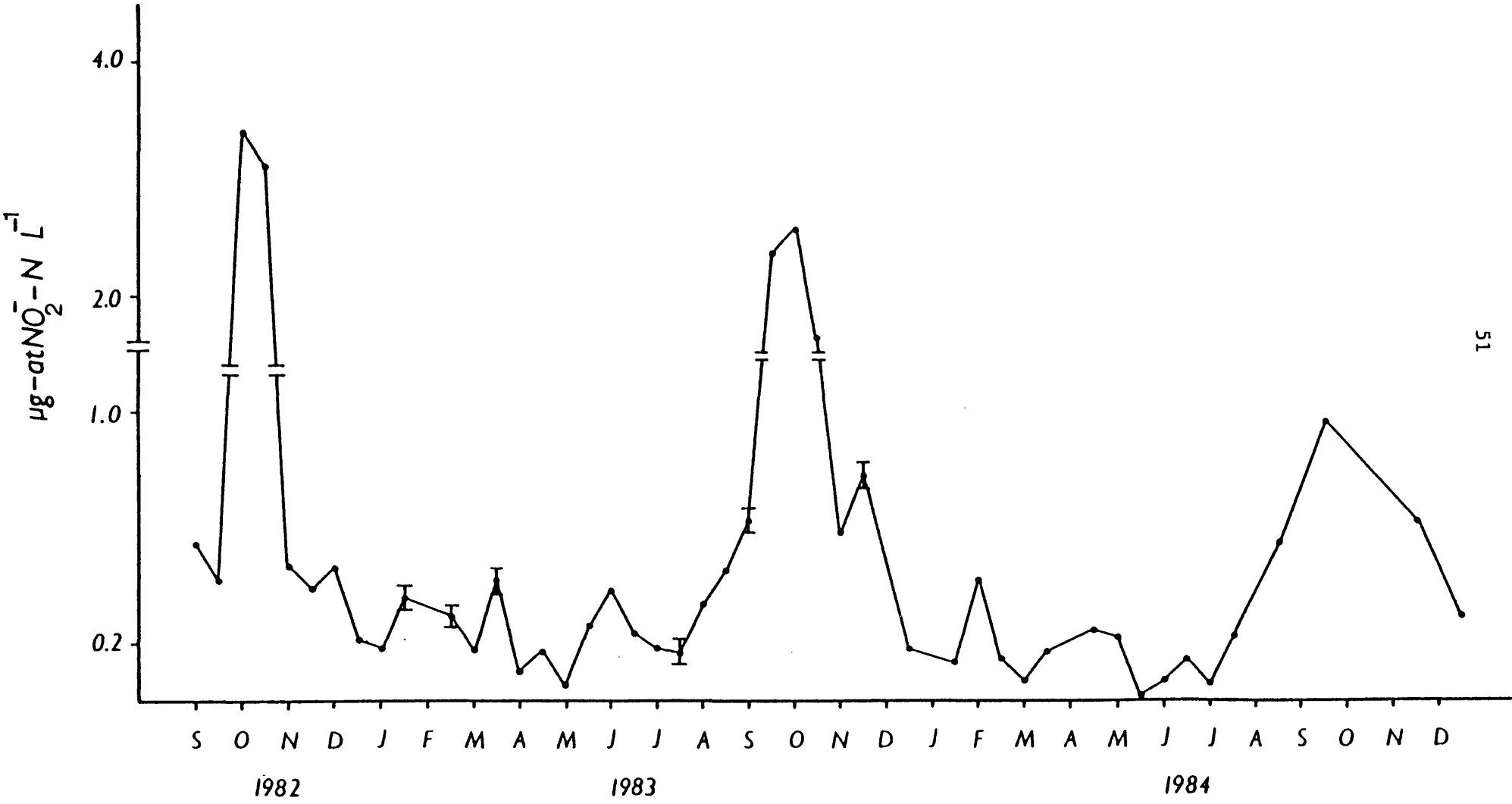
sudden increase to ca. 3.32 µg at 1^{-1} in October, 1982, following which the concentration dropped to 0.39 µg at 1^{-1}. During the following period the concentration remained around 0.25 µg at 1^{-1} until it reached its minimum concentration of 0.075 µg at 1^{-1} in July, 1983. This was followed by a rapid increase up to the highest maximum concentration of ca. 2.6 µg ay 1^{-1} October, 1983, followed by a drop in the concentration until it reached 0.18 µg at 1^{-1} in December. Nitrite concentration remained around this level until May, 1984 when it reached its minimum of ca. 0.019 µg at 1^{-1}. Thereafter, there was a steady increase up to ca. 1 µg at 1^{-1} in September, 1984.

ii. Station B

In this station nitrite concentration was observed over a range of ca. 0.019-3.4 µg at 1^{-1} (Fig. 3.10).

There was a sudden increase from the September, 1982 concentration of 0.31 µg at 1 up to 3.4 µg at 1^{-1} in October. The concentration dropped sharply down to 0.475 µg at 1^{-1} in November. There was a slow decrease from the November concentration of 0.475 µg 1^{-1} down to 0.062 µg at 1^{-1} in May, 1983. This was the minimum value observed in the 1982-1983 period. This value was followed by a rapid increase up to 2.56 µg at 1^{-1} in October. A decrease in the concentration down to ca. 0.19 µg at 1^{-1} in December was observed. The concentration remained around this level for the following months until May, 1984 when it reached a minimum of ca. 0.019 µg at 1^{-1}. This was followed by a rapid increase up to 0.96 µg at 1^{-1} in September, 1984.

Figure 3.10 Nitrite-nitrogen (Station B)


4.0
2.0
1.0
0.2
µg-at NO_2^--N L^{-1}
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

8. Ammonia-nitrogen

i. Station A

Ammonia concentration was relatively high when the sampling programme started in September, 1982, with a peak of ca. 0.49 µg at l^{-1}. The concentration values which followed were usually between 0.05 and 0.25 µg at l^{-1} (Fig. 3.11). These values characterized a period extending from October, 1982 to May, 1983. In June, 1983 a minimum value of 0.014 µg at l^{-1} was observed. This point represented the end of an annual cycle and the start of a new cycle by the steady increase of the concentration until it reached a high concentration of ca. 0.28 µg at l^{-1} during the months from August to October, 1983. These months of relatively high ammonia concentration were followed by a period of relatively low concentrations (0.1 µg at l^{-1}). This period was followed by a drop down to a minimum concentration of 0.008 µg at l^{-1} in May, 1984. Once again the concentration started to rise steadily until it reached a value of ca. 0.3 µg at l^{-1}. This was followed by a drop down to 0.058 µg at l^{-1} in December.

ii. Station B

In this station, the ammonia concentrations showed variable values throughout the year (Fig. 3.12), but in general the values during the winter-early spring months were around or above 0.1 µg at l^{-1} especially in 1982-1983.

These relatively high concentrations were followed by a drop down to a summer minimum (0 µg at l^{-1}). Throughout this study the high peaks which were observed during the autumn-spring periods had values around 0.3-0.45 µg at l^{-1}. From these high peaks, two were exceptional, one in November, 1982 (0.64 µg at l^{-1}) and another in February, 1984 (0.58 µg at $^{-1}$).

Figure 3.11 Ammonia-nitrogen (Station A)

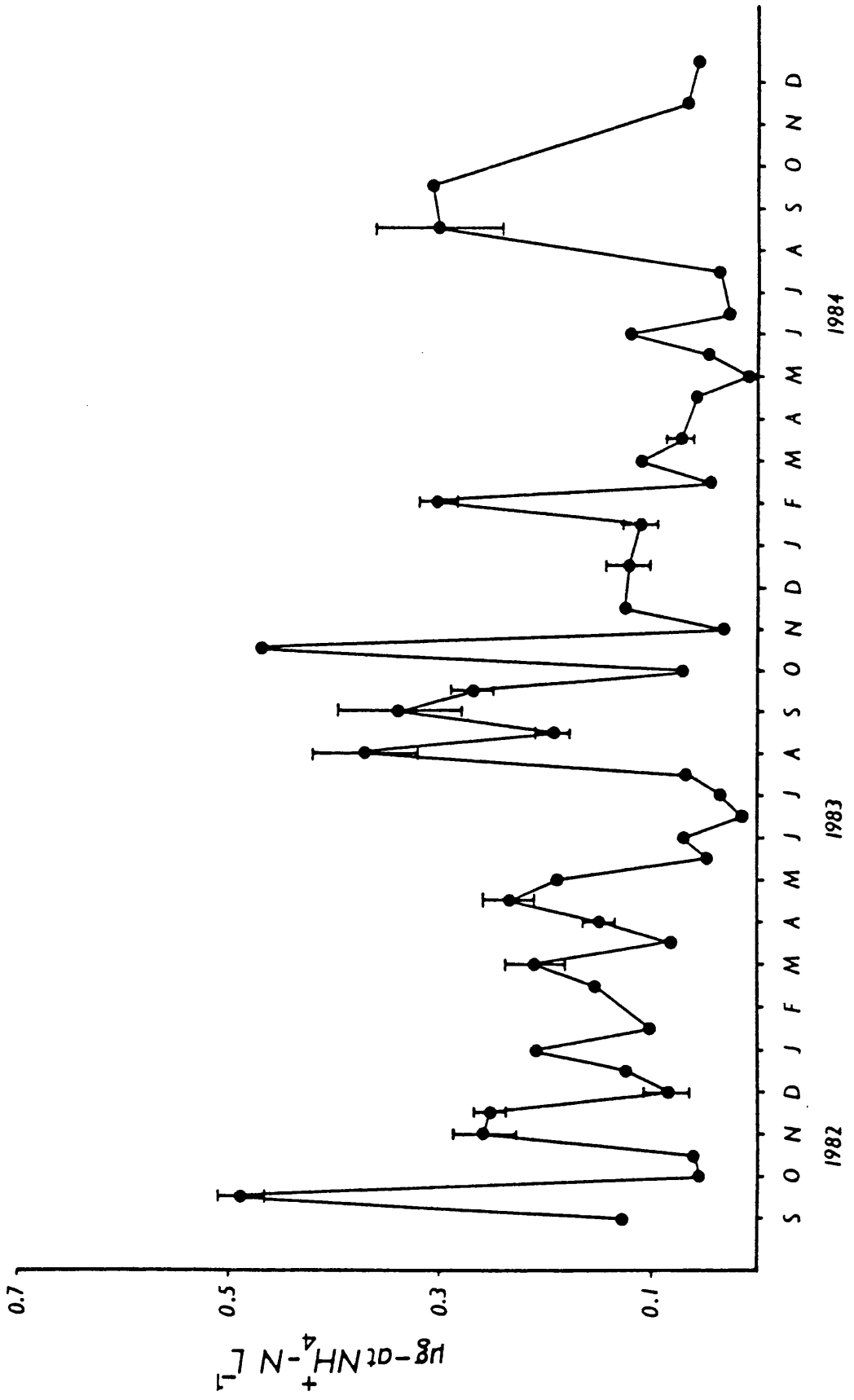
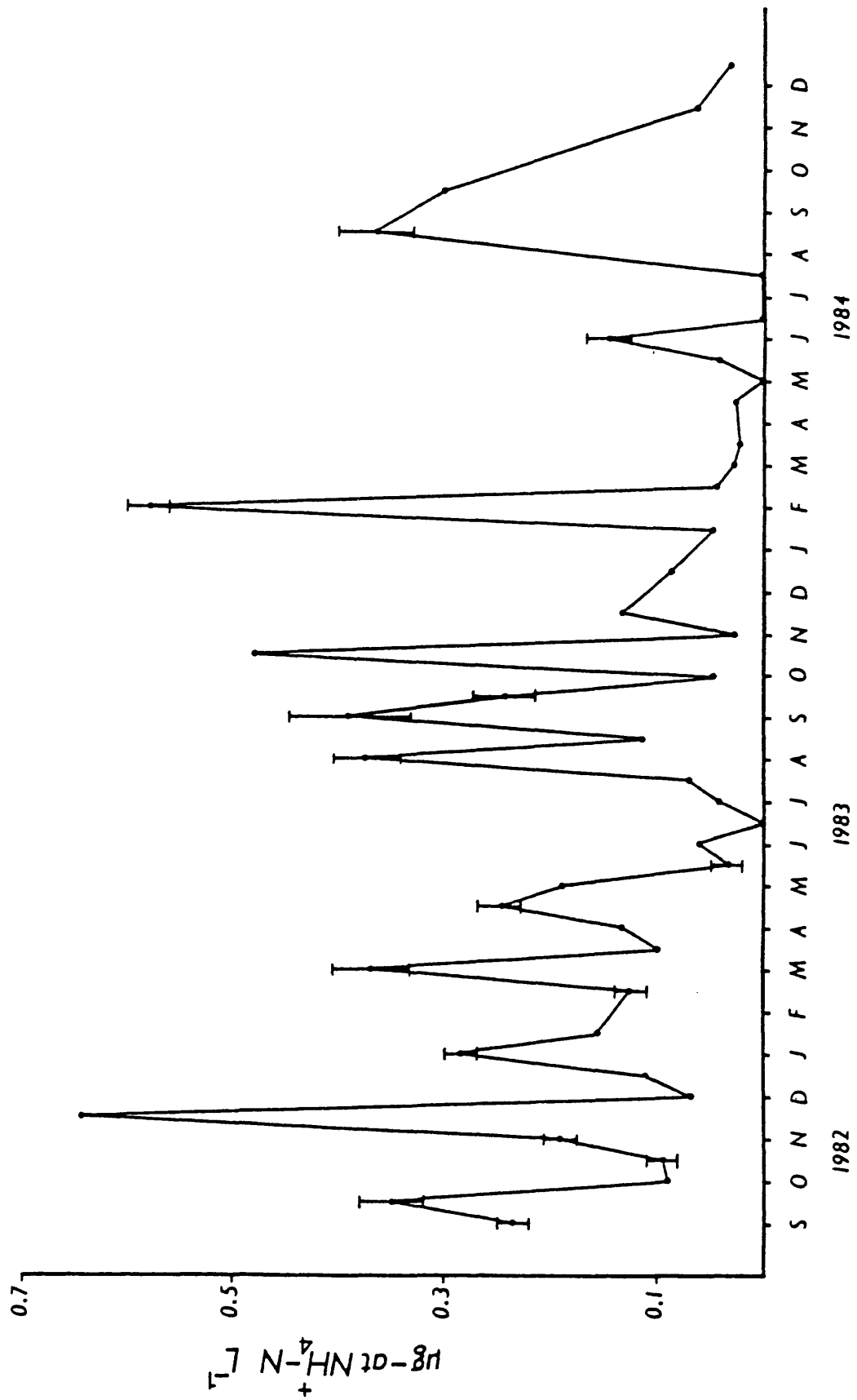

0.7
0.5
0.3
0.1
µg-at NH4-N L-1
S O N D J F M A M J J A S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

Figure 3.12 Ammonia-nitrogen (Station B)


0.7
0.5
0.3
0.1
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

9. Phosphate-phosphorus

i. Station A

Phosphate-phosphorus concentration was observed over a range of 0.058-1.8 μg at l^{-1} (Fig. 3.13).

The phosphate concentration in September, and October, 1982 of ca. 0.5 μg at l^{-1} increased rapidly during the following month up to ca. 1.8 μg at l^{-1} (November, 1982). During the following months until May, 1983, the concentrations were around 1 μg at l^{-1}. A sudden drop from this level down to 0.06 μg at l^{-1} was observed in July, 1983.

At the end of July, the concentration increased up to 0.87 μg at l^{-1} and remained above 0.4 μg at l^{-1} for the period until May, 1984. During this period of high concentration, a maximum value of ca. 1.8 μg at l^{-1} was observed in February, 1984. During the period from May to July, 1984 the concentration remained at the low level of ca. 0.17 μg at l^{-1}. By early August, 1984 the concentration started to increase up to 1.45 μg at l^{-1} in December.

ii. Station B

The phosphate-phosphorus concentrations in this station were observed over a range of 0.06-2.26 μg at l^{-1} (Fig. 3.14).

At the start of this study, phosphate concentration was ca. 0.54 μg at l^{-1} in September, 1982. It increased after September and remain at a high level of ca. 1 μg at l^{-1} until June, 1983. An exception to the high autumn-spring concentrations, was that observed in March, 1983. In that month, a very low value of 0.12 μg at l^{-1} was found.

A sudden drop from the high concentration of early June, 1983 down to a minimum concentration of 0.064 μg at l^{-1} was observed in June-July. This low concentration remained for a short period and was followed by

Figure 3.13 Phosphate (Station A)

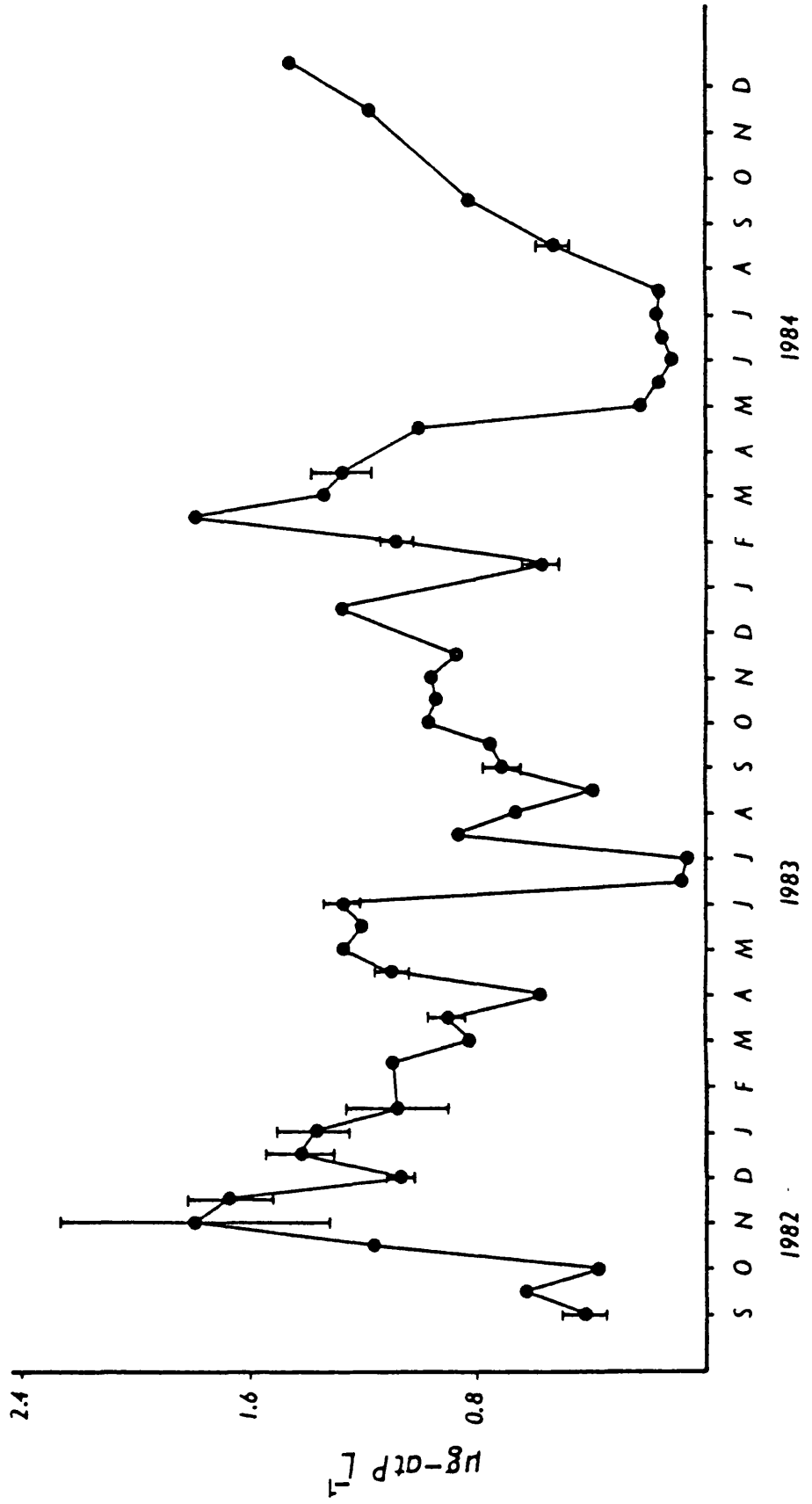
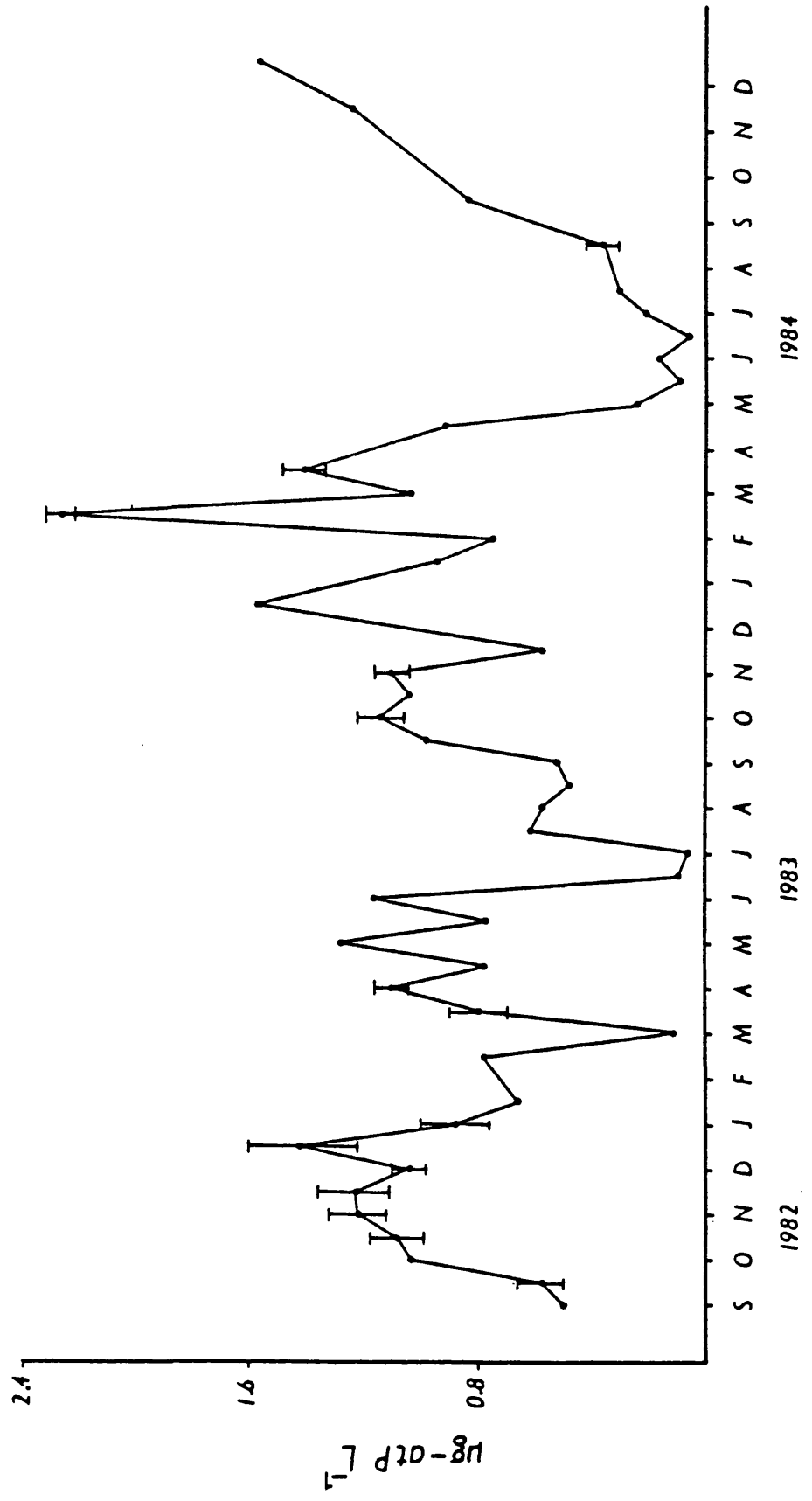

µg-atP L⁻¹
2.4
1.6
0.8
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

Figure 3.14 Phosphate (Station B)


µg-atP L⁻¹
2.4
1.6
0.8
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

a rapid increase in late July. During the following period from August, 1983 to April, 1984, the concentrations remained high and most of the time above 1 µg at l^{-1}. During this period, an exceptionally high concentration of phosphate was observed in February, 1984 (2.26 µg at l^{-1}). The period of low phosphate concentrations extended from May to July, 1984 with values around 0.17 µg at l^{-1} and a minimum one of 0.06 µg at l^{-1} in June. Thereafter, the concentration increased rapidly reaching a value of 1.57 µg at l^{-1} in December, 1984.

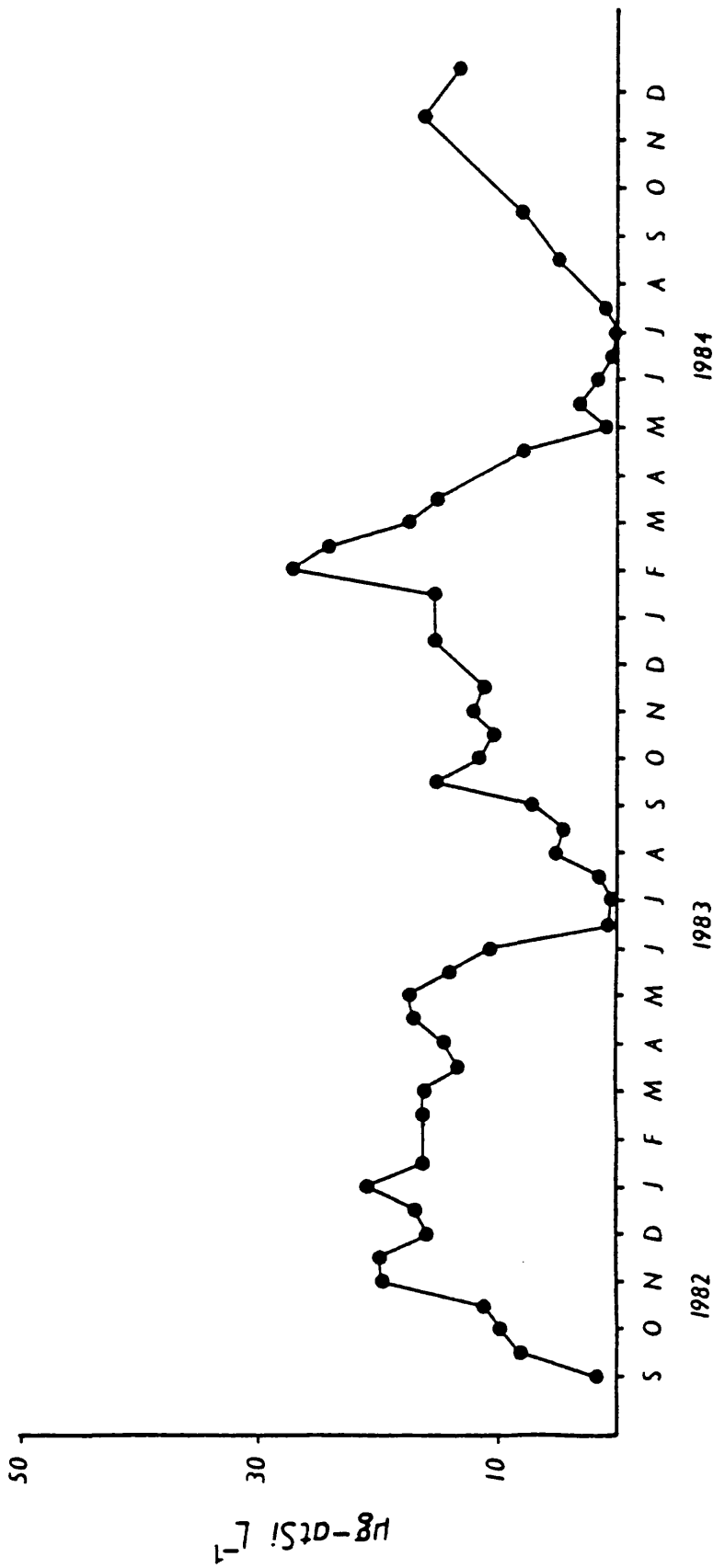
10. Silicate-silicon

i. Station A

Silicate concentration was observed over a range of 0-27.21 µg at l^{-1}) (Fig. 3.15).

The low silicate concentration of September, 1982 (1.73 µg at l^{-1}) was followed by an increase up to 19.96 µg at l^{-1} in early November, 1982. The silicate concentration remained around this high level until June, 1983 when there was a drop down to 0.51 µg at l^{-1} in July. This was followed by a steady increase up to ca. 15 µg at l^{-1} in September. The concentrations remained around this level until January, 1984. In February, a sudden increase up to 27.21 µg at l^{-1} was observed. This was followed by a steady decrease down to the low summer level in May, 1984. The concentration remained at this low level until it reached a minimum of ca. 0 µg at l^{-1} in July. This minimum concentration was followed by a steady increase up to ca. 16.5 µg at l^{-1} in November, 1984.

Figure 3.15 Silicate (Station A)


µg-at Si L⁻¹
50
30
10
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

ii. Station B

The silicate-silicon concentration observed in this station was over a range of 0-42.53 µg at l^{-1} (Fig. 3.16).

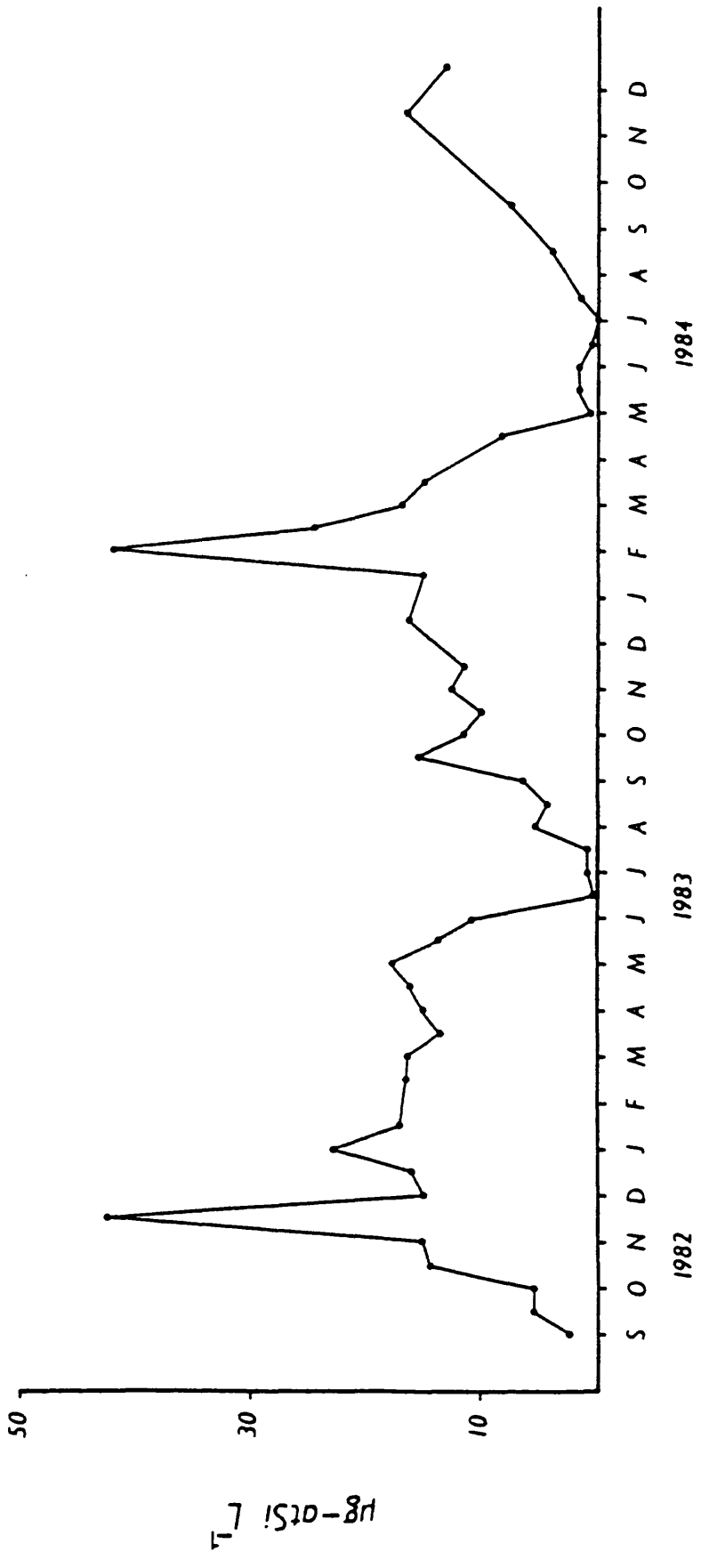
The relatively low concentration of September, 1982 (2.55 µg at l^{-1}) was followed by a rapid increase up to the highest maximum of 42.53 µg at l^{-1} in November. The concentration dropped from this maximum down to ca. 15 µg at l^{-1}. This relatively high concnetration was observed over the period extending from December, 1982 to June, 1983. By late June, the concentration reached ca. 0.5 µg at l^{-1}. This remained until late July when it started to increase rapidly up to ca. 15 µg at l^{-1} in September. The concentration remained at that level until January, 1984. In February, a sudden increase up to ca. 42 µg at l^{-1} was observed. This was followed by a steady decrease in the concentration down to ca. 0.85 µg at l^{-1} in May, 1984. The concentration remained at this low level reaching a minimum of 0 µg at l^{-1} in July. Thereafter, the concentration increased rapidly reaching ca. 16.5 µg at l^{-1} in November, 1984.

DISCUSSION

The physical and chemical oceanography of Swansea Bay have been studied by many authors (Paulraj, 1974; Chubb et al, 1980; Humphrey et al, 1980; Joint, 1980; Morris and Mantoura, 1980).

The data of Paulraj (1974) showed a clear seasonal variation in silicate. Phosphate concentration was less variable, and nitrate concentration did not show any seasonal trend. Morris and Mantoura (1980) studied the water chemistry of outer Swansea Bay during five

Figure 3.16 Silicate (Station B)


µg-atSi L⁻¹
50
30
10
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

cruises between February, 1977 and January, 1978. They reported a 100% depletion in the silicate concentration during the summer months, while total oxidised nitrogen and orthophosphate were depleted to some 30-40% of winter values.

Humphrey et al. (1980) have discussed the distribution of nutrients throughout the Bay, using data collected by the Welsh Water Authority at four tidal states on each of six occasions during 1976/1977. They found that the nutrient concentrations in winter are higher than in most coastal regions of the Bristol Channel. A well-defined seasonal variation in silicate concentrations has been found. They pointed out that the seasonal variations of phosphate and nitrate in the inshore waters were masked to a certain extent by the effect of nutrient input. Chubb et al. (1980) estimated that approximately 9.75 tonnes of inorganic nitrogen are discharged daily to the bay. The daily orthophosphate input was about 729 kg day^{-1} and the silicate input was 13 tonnes day^{-1}.

However, the present study showed clearly a well-defined seasonal variation of the major nutrients (nitrate, nitrite, phosphate and silicate). There was an increase of nitrate up to ca. 49 µg at l^{-1} (2.11., 1982, A) and 66 g at l^{-1} (17.2., 1984, A). The concentration of nitrate in the other station (B) reached the same level during the autumn-winter period. At both stations, nitrate was depleted to 1% of the winter level during the two annual cycles recorded.

Nitrite concentrations were depleted to ca. 2% of the winter maxima. This depletion occurred at both stations after each winter maximum was reached. The same trend was observed for phosphate and silicate. During the 28 months of sampling, the winter phosphate maxima dropped in the summer down to 2-6%.

The summer silicate concentrations represented 0-2% of the winter maxima. The data of this study show very different concentrations of the major nutrients (especially the nitrate and phosphate) than those recorded by Paulraj (1974), Humphrey et al. (1980), and Morris and Mantoura (1980).

It is very obvious that the seasonal variations of nitrate, nitrite, phosphate and silicate were not masked by the domestic and industrial discharge into Swansea Bay (Humphrey et al. 1980). It seems that the results obtained by Morris and Mantoura (1980), and Humphrey et al. (1980) do not give the true image of nutrients' seasonal variations in Swansea Bay. The data of these two researchers were collected during selected cruises, and it is very likely that periods of significant nutrient concentrations have been missed. This could be the reason why well-defined seasonal variations of phosphate and nitrate were not found. This explanation is based on the results of the present study. It can be seen from the nutrient data (Figs. 3.7-3.16) that very low nutrients concentration occurred during a short period. For example, if the period of July, 1983 was missed, the lowest nitrate value of Station A would be 4.4 μg at l^{-1} which is ca. 9% of the winter maximum of that annual cycle. But because the July period was sampled, the minimum value of nitrate concentration found was 0.78% and this can be applied to the other nutrients. The results of the present study emphasize the role of nutrients (including nitrate) as important factors in Swansea Bay, and their possible role as limiting factors should not be underestimated.

The continuous variation in ammonia concentration in the present study makes it difficult to give a reliable explanation for the source or fate of ammonia and its effect on the primary productivity of

Swansea Bay.

Although there was a trend towards seasonal variation (Figs. 3.11 and 3.12), the ammonia concentration does not correspond to phytoplankton growth. This indicates that the effect of ammonium uptake by phytoplankton on seasonal change of ammonia concentration was minimal. A possible source of ammonia variation may have been animal excretion, in addition to industrial discharges into Swansea Bay, especially along the north-eastern coastline (W.W.A., 1983).

In the biological communities nitrate is reduced to ammonium with nitrite as intermediate form and vice-versa. In the present study, the inorganic nitrogen sources have shown seasonal maxima at different times of the year. In the winter, especially that of 1982-1983, ammonia maximum took place first, followed by nitrite and then nitrate. This order of occurrence indicates that there were oxidation processes taking place in that period of the year. This suggestion is further supported by the occurrence of exceptional nitrite maxima in October, 1982, and October, 1983, coinciding with a drop in ammonia concentration.

Brandhorst (1958, 1959) suggested that a nitrite peak in the natural enviroment was due to bacterial oxidation of ammonia. This hypothesis was supported by the observation that an increased production of nitrite has been found when samples containing added ammonium are maintained in the dark (Wada and Hattori, 1971; Hattori and Wada, 1972; Miyazaki et al., 1973).

Air and seawater temperatures exhibited a similar pattern, which follows the seasonal cycle of temperature in the temperate regions (Sverdrup et al., 1942). Their strong correlation indicates that they have a common source of heat energy, which is solar radiation.

Temperature plays an important role in the primary productivity of phytoplankton and this role becomes significant when it is coupled with long periods of sunshine hours (Sykes, 1981). In Swansea Bay, the coupling of long periods of sunshine and temperature occurred mainly during the spring and summer months. On the other hand, the winter months were characterized by short periods of sunshine accompanied by low temperatures (Figs. 3.1, 3.2 and 3.3). These two factors may have been limiting to the growth of phytoplankton in the winter. The strong winter gales accompanied by the strong tides, characteristic of Swansea Bay (and the Bristol Channel as a whole), increase the degree of vertical mixing and hence the turbidity. The high turbidity decreases the penetration depth of sunlight, and when coupled with short sunshine periods and low temperature it could play a very important role in limiting phytoplankton growth in winter (Joint and Pomroy, 1981).

The very low salinities observed in November, 1982, and February, 1984 (Fig. 3.6) were preceded by high levels of rainfall. At the same time, exceptionally high levels of silicate were found (ca. 42 μg at Si l^{-1}: Fig. 3.16). These observations were noticed at the western station (B), where the input of the River Neath could be most effective. Chubb et al. (1980) found that the major silicate input to Swansea Bay was through the rivers (86.7%). In the same study, the River Neath has been found to represent the highest percentage of the major input total (27.7%). High levels of silicate were found in February, 1984 at the other station (A) but to a lesser degree (27 μg at Si l^{-1}).

The anticlockwise circulation described by Collins et al. (1979) and supported by the salinity data of Humphrey et al. (1980), who found that fresh water from the Rivers Neath and Tawe dispersed in a

southerly/south-westerly direction has been used as an explanation of the very low salinities found in the area where the two rivers discharge (Humphrey et al., 1980). The low salinity and high silicate concentration values found at Station (B) (which is nearer to the area of eddy circulation) can be explained by the presence of the anti-clockwise eddy circulation. The effect of river input on the salinity and silicate values is supported by the strong correlation between salinity and silicate ($r = -0.69$ (A), and $r = -0.813$ (B)).

Nitrate and phosphate correlate less strongly with salinity than does silicate (Table 3.1). The correlation between salinity and the major plant nutrients suggests that river runoff was the main controlling factor in the distribution of nutrients in Swansea Bay. The effect of the major environmental factors on the biomass (estimated as chlorophyll a concentration) was investigated by determining the correlation coefficient between chlorophyll a and each of the factors. (Data were omitted when there was suspected limitation by any factor other than the one under investigation.) The correlation coefficients are listed in Table 3.1. From the correlation coefficient results it can be seen that at Station (A) nitrate and silicate were correlated with chlorophyll a. At this station, chlorophyll a was strongly correlated with sunshine hours. On the other hand, at Station (B) silicate was the only nutrient which was correlated to a limited extent with chlorophyll a. The lack of strong correlation between chlorophyll a and all the measured environmental factors at Station (B) suggests that production at this station may have been controlled by a local factor not recorded in this study.

The strong correlation at Station (A) between chlorophyll a and sunshine hours suggests that the later had a main role in limiting the

Table 3.1

FACTOR	CORRELATION COEFFICIENT	SIGNIFICANT AT P >
Salinity v nitrate (A)	- 0.48	0.005
Salinity v nitrate (B)	- 0.425	0.005
Salinity v phosphate (A)	- 0.434	0.005
Salinity v phosphate (B)	- 0.264	0.005
Salinity v silicate (A)	- 0.69	0.005
Salinity v silicate (B)	- 0.813	0.005
Nitrate v chlorophyll *a* (A)	- 0.3329	0.005
Nitrate v chlorophyll *a* (B)	- 0.1028	0.005
Phosphate v chlorophyll *a* (A)	- 0.1624	0.005
Phosphate v chlorophyll *a* (B)	+ 0.013	0.005
Silicate v chlorophyll *a* (A)	- 0.44	0.005
Silicate v chlorophyll *a* (B)	- 0.249	0.005
Sunshine hours v chlorophyll *a* (A)	+ 0.763	0.005
Sunshine hours v chlorophyll *a* (B)	+ 0.26	0.005

production during winter periods. While the correlation between chlorophyll *a* and the plant nutrients at the time when the average daily sunshine hours was > 2 indicates that these nutrients may have been limiting to the production in the spring and summer periods.

CHAPTER IV

ABUNDANCE, BIOMASS AND PRIMARY PRODUCTIVITY

INTRODUCTION

Ecologically significant organisms are those which play an effective role in the ecology and dynamics of an ecosystem. The significant status of phytoplankters is derived from their role as primary producers in the aquatic environment.

Several methods and techniques have been used for the evaluation of phytoplankton abundance, biomass and primary productivity. Although direct counting of phytoplankton has been used for a long time, its results can not be used as representative of the biomass because phytoplankton differ greatly in size. In fact the main advantage of this method is its use as a tool in the qualitative differentiation between species and differentiation of organisms from detrital particles. Cell volume, weight of the available matter per volume of water and carbon content of the plant have all been used as indicators of phytoplankton biomass.

Perhaps one of the most universally used techniques for the determination of biomass is the calculation of chlorophyll a content. The method which is widely used is the one developed by Strickland and Parsons (1972).

Primary productivity is another parameter which has been thoroughly investigated in the aquatic environments. Gaarder and Gran (1927) gave an account on the technique which involves the calculation of the change in oxygen production. Measurement of the radioactive carbon uptake by the photosynthetic organisms is another widely used technique for the estimation of primary productivity.

The interactions of zooplankton and phytoplankton raised the possibility that the zooplankton could be one of the environmental

factors which may concurrently or sequentially influence the growth of natural populations of phytoplankton (Platt et al., 1977).

Plankton ecology and production in Swansea Bay and South Gower coastal waters have been studied by Pearce (1967), Gabriel (1973), Issac (1974), Paulraj (1974), Tyler (1976), Joint (1980) and Vogelmann (1980).

Paulraj (1974), who studied the phytoplankton at Mumbles pier, found an annual fluctuation in chlorophyll a concentrations from 1.7 mg m^{-3} in July, 1973 to 26.6 mg m^{-3} in May, 1974. Joint (1980) studied the spatial variability in a grid just outside Swansea Bay during August, 1977. He found that at some stations chlorophyll a concentrations were higher than at others, reaching a maximum of about 6 mg m^{-3}. In the present study, different parameters were used to study the phytoplankton production in Swansea Bay. These were cell number, chlorophyll a concentration and uptake of radioactive carbon.

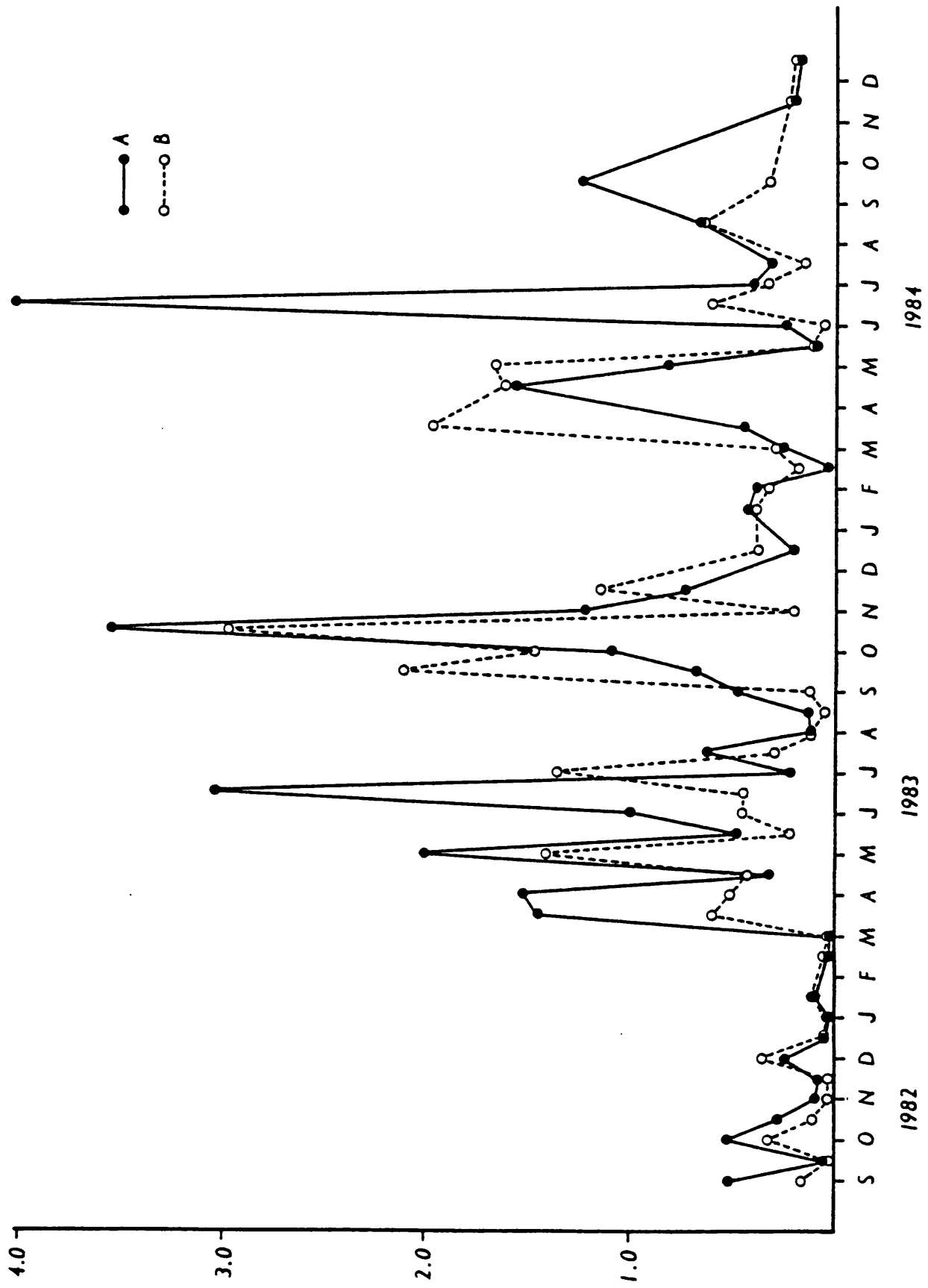
RESULTS

I. Phytoplankton Annual Cycles

a. Total number of net phytoplankton:

In general, the total cell number of net phytoplankton for both stations follows the general pattern of seasonal variation in temperate regions (Fig. 4.1). In the winter of 1982-1983 the number of cells was at its lowest value. This was followed by several peaks during March to June, 1983. After a period of low cell number in July and August, 1983, high peaks occurred in September and October, 1983. This was followed by gradual drop with a minimum value in February,

Figure 4.1 Total phytoplankton cell number


Cell number X10³ m⁻³
4.0
3.0
2.0
1.0
A
B
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

1984. The following spring was characterized by the occurrence of relatively high peaks of phytoplankton. A striking result was seen in June, 1984 when the cell number in Station A was at its maximum for the whole period of the study, but at the same time Station B showed a relatively low number of cells. There was a slight increase in the cell number in August and September, 1984 which followed a drop in the cell number during July. This increase was more obvious in Station A than in Station B.

In general, it can be said that there were some significant differences between the total number of net phytoplankton of Station A and that of B. Examples of such differences are shown in Table 4.1

Table 4.1 Total number of phytoplankton

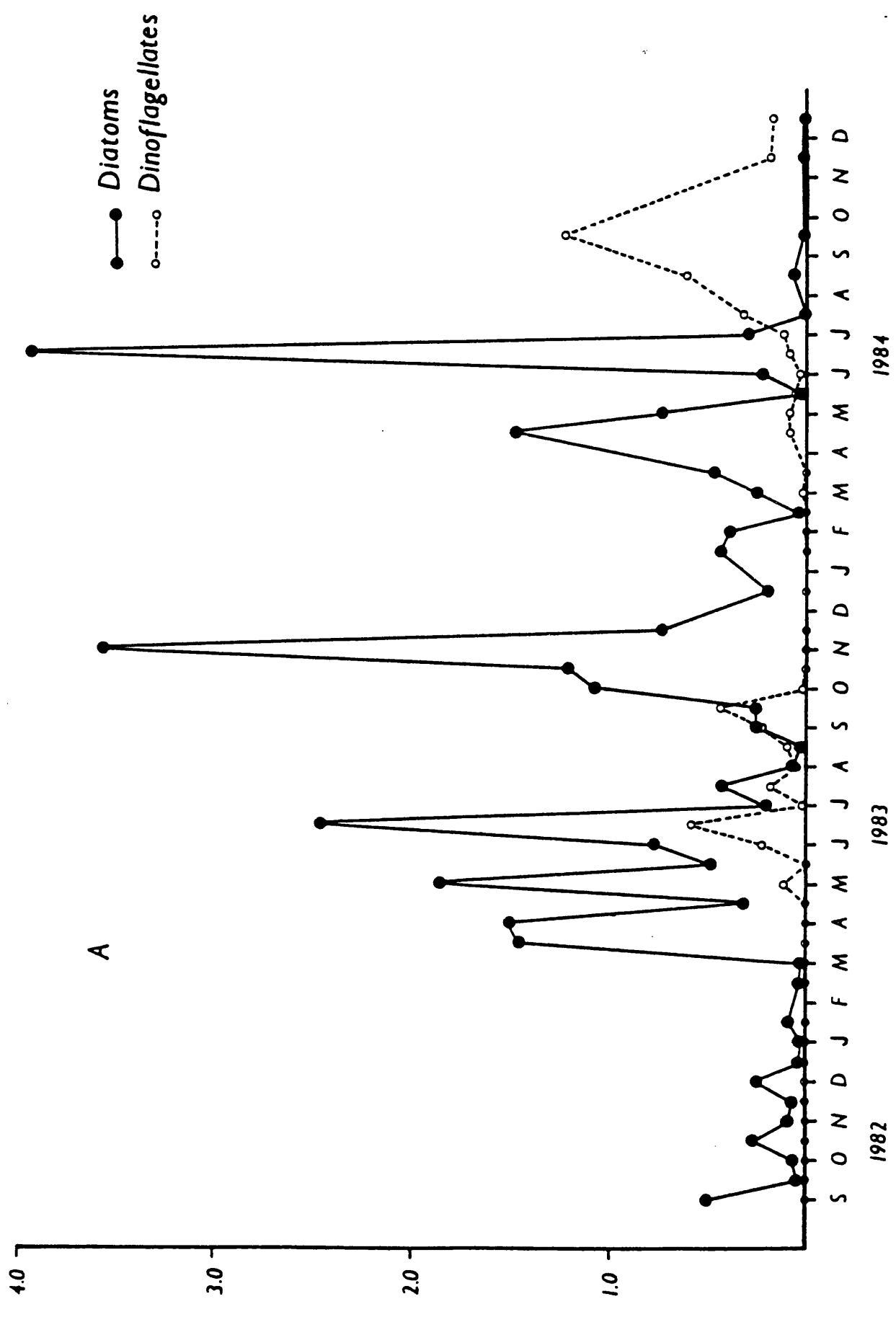
DATE	cell m^{-3} (A)	cell m^{-3} (B)
31. 3. 1983	1462	599
12. 4. 1983	1517	505
23. 6. 1983	2475	375
22. 9. 1983	688	2112
20. 3. 1984	463	1976
27. 6. 1984	4027	607

b. Phytoplankton groups

i. Station A:

In the temperate region, diatoms represent the major group in the marine phytoplankton. In Station A (Fig. 4.2), the high peaks of phytoplankton which occurred during the seasons of rapid growth consisted mainly of diatoms. This was recorded in March, April, May

Figure 4.2 Diatoms and dinoflagellates
(Station A)


Cell number X 10³ m⁻³
4.0
3.0
2.0
1.0
A
Diatoms
Dinoflagellates
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

and November, 1983. Over 90% of the net plankton cells in the April and June peaks were dominated by diatoms.

In general, the maximum peaks of flagellated phytoplankton were usually lower than the maximum peaks of diatoms. Dinoflagellates blooms occurred at the end of, and after, the diatom spring bloom of 1983. The highest peak of flagellates took place in September, 1984.

ii. Station B:

During the spring and summer of 1983, two diatom peaks occured in May and July (Fig. 4.3). The highest peak of diatom in 1983 was seen in October followed by a smaller one in November. The maximum diatom bloom in 1984 occurred in March, April and May. A smaller peak occurred in June and another one in August. The general picture of flagellates growth during 1983 was dominated by relatively high peaks from May to June, and another peak in September. The highest bloom of flagellates in 1984 took place in August, and gradually decreased after that. As in Station A, the peaks of flagellates were generally smaller than those of the diatoms.

II. Chlorophyll a

i. Station A:

A high value of 14.27 mg m^{-3} was found in September, 1982 (Fig. 4.4). This was followed by a drop to 1.955 mg m^{-3} which continued until April, 1983 when chlorophyll a concentrations started to increase. It reached a maximum of ca. 52 mg m^{-3} in June when then it dropped. The low concentration was maintained until April, 1984 with the exception of a small rise in August, 1983. In the spring of 1984 it reached a maximum of ca. 16 mg m^{-3} in May. After reaching the maximum it

Figure 4.3 Diatoms and dinoflagellates
(Station B)

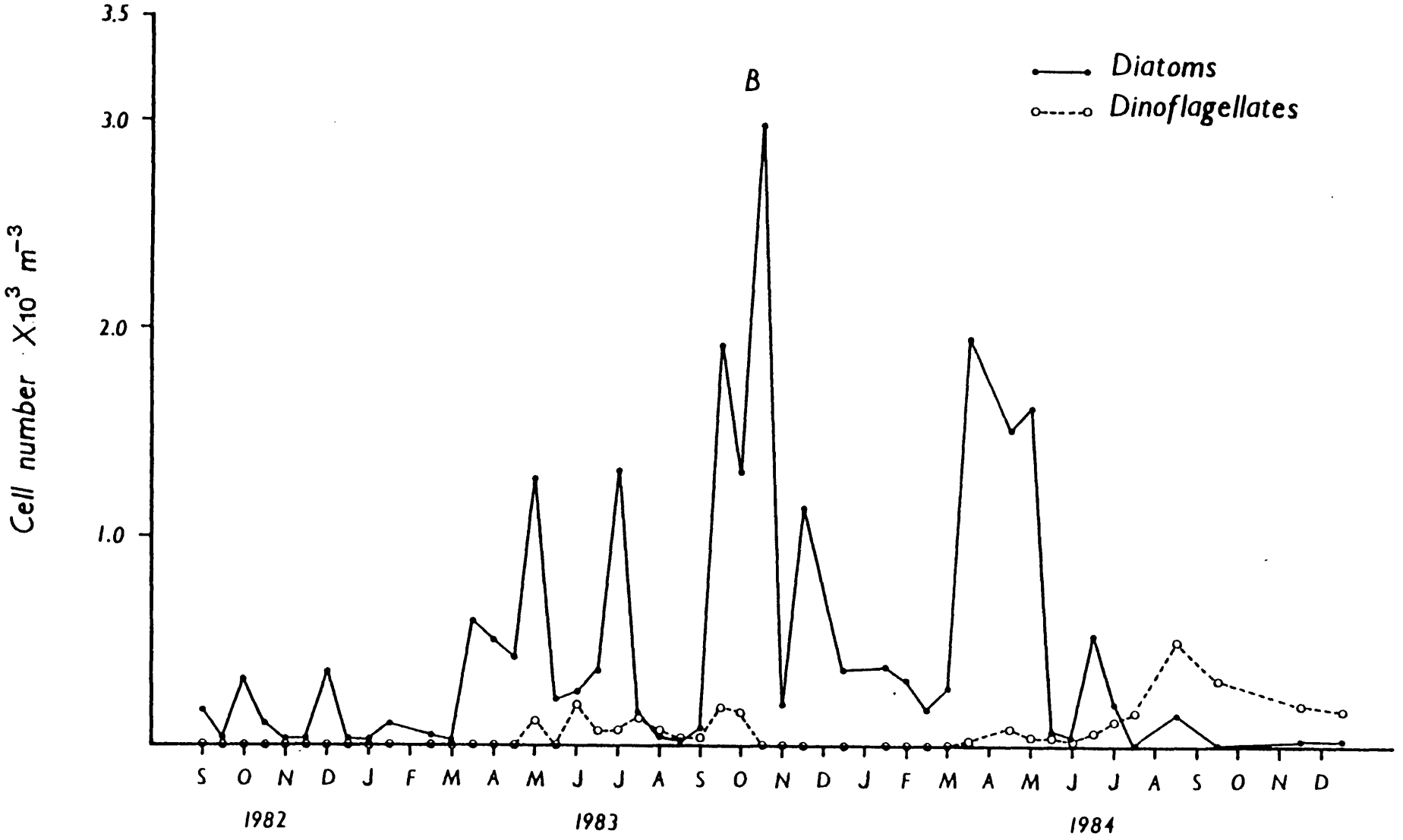
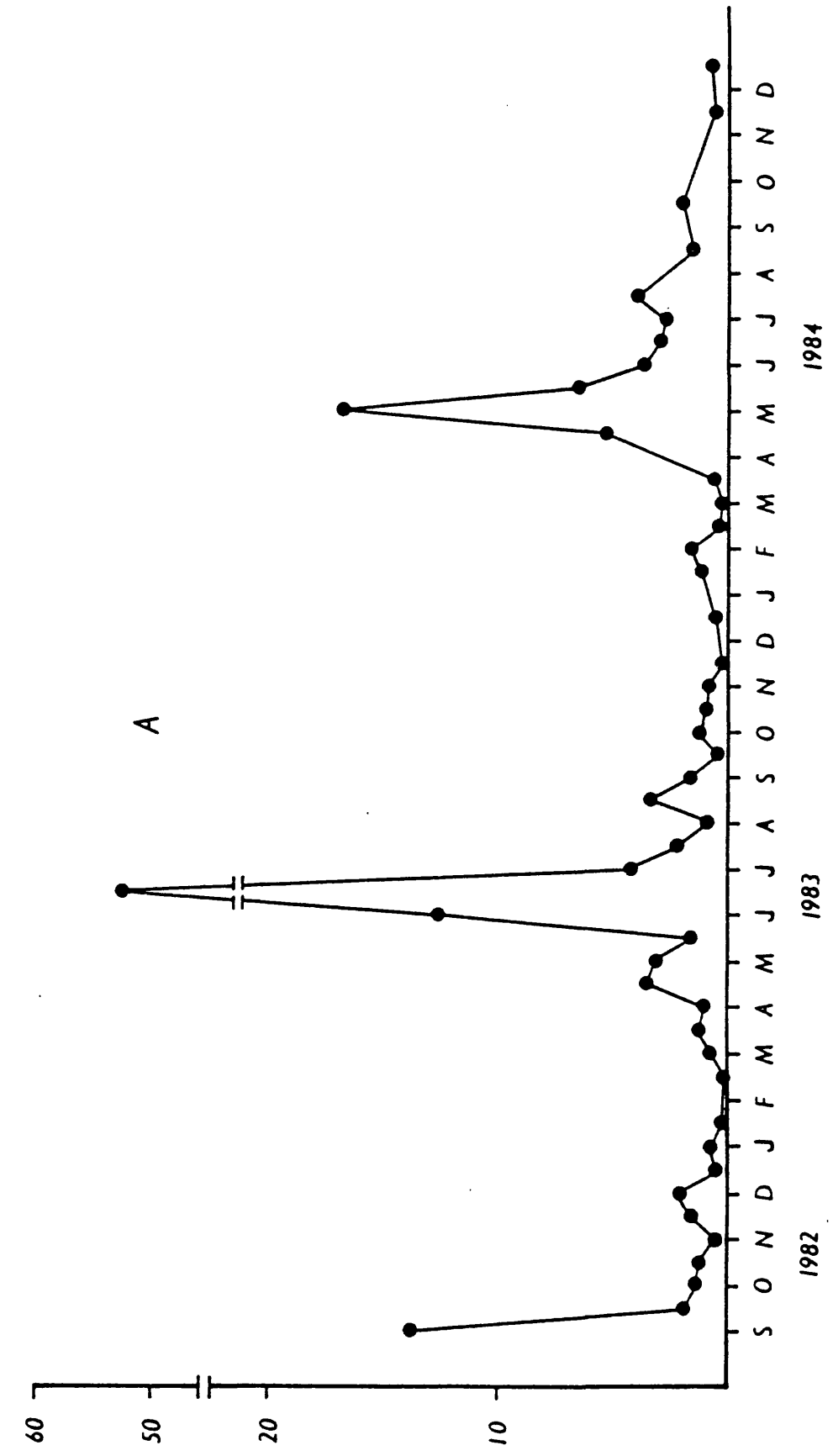

3.5
3.0
2.0
1.0
Cell number X10³ m⁻³
B
Diatoms
Dinoflagellates
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

Figure 4.4 Chlorophyll *a*
(Station A)


mg chlorophyll a m⁻³
60
50
20
10
A
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

gradually dropped reaching a minimum of 0.65 mg m^{-3} in November, 1984.

ii. Station B:

Almost the same pattern of variation observed in Station A was found in Station B (Fig. 4.5). A value of ca. 7 mg m^{-3} in September, 1982 was followed by a gradual decrease until April, 1983 when it started to increase. It reached a maximum of ca. 50 mg m^{-3} in June, 1983 followed by a small rise in August. The low values during the following months remained until April, 1984 when it started to increase reaching a maximum value of ca. 16 mg m^{-3} in May. The concentration of chlorophyll a remained at a value of ca. 5 mg m^{-3} from the end of May, 1984 until late July when it dropped and remained low.

III. Primary Productivity of Phytoplankton

i. Station A:

The fluctuations in the values of primary productivity for Station A followed the pattern of chlorophyll a concentration seasonal change (Fig. 4.6). A value of ca. 14 mg C m^{-3} hr^{-1} obtained in September, 1982 was followed by a sudden drop to a low value of ca. 1 mg C m^{-3} hr^{-1}. This low value remained almost constant until April, 1983. The productivity started to increase from April reaching a maximum of ca. 51 mg C m^{-3} hr^{-1} in June, 1983. The productivity fluctuated around a level of ca. 8 mg C m^{-3} hr^{-1} from August to November, 1983. A very low value of ca. 0.6 mg C m^{-3} hr^{-1} in late November remained at the same level until April, 1984. A gradual increase started in April reaching a maximum of ca. 56 mg C m^{-3} hr^{-1} in May followed by a drop in June. A relatively high peak of ca. 12 mg C m^{-3} hr^{-1} was found in July, 1984. The productivity then started to decrease until the end of the sampling

Figure 4.5 Chlorophyll *a*
(Station B)

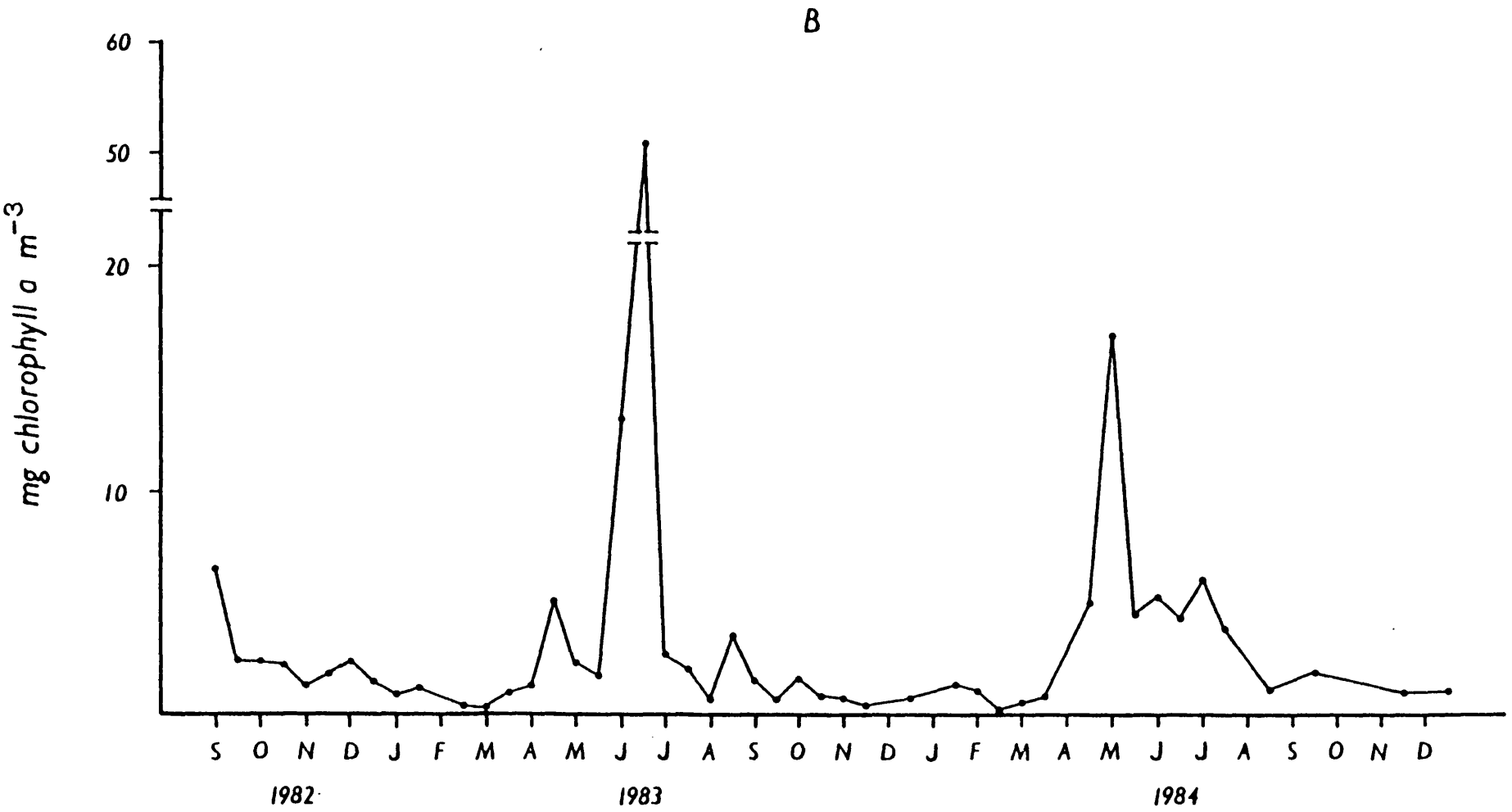
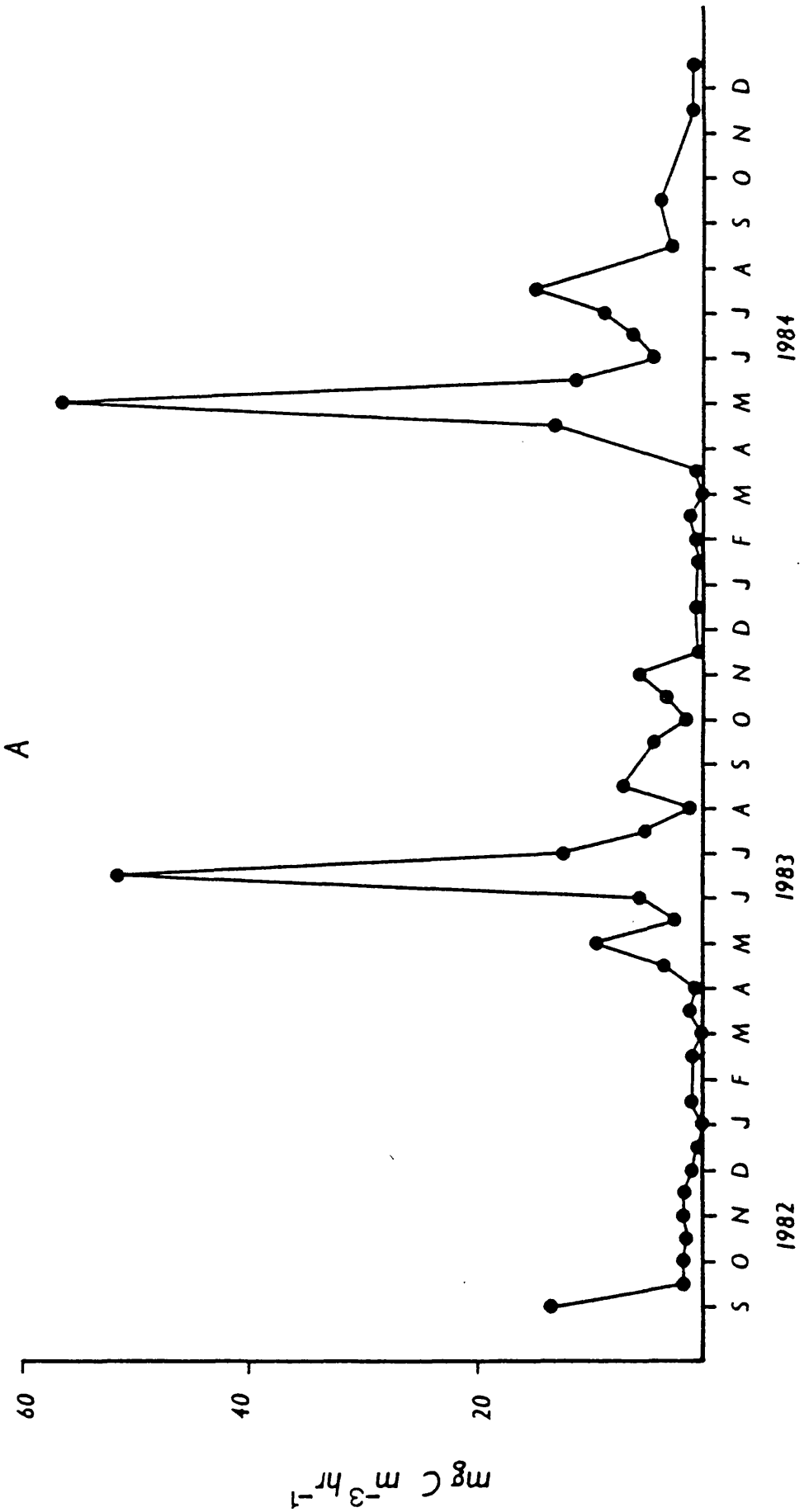

B
60
50
20
10
mg chlorophyll a m^{-3}
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

Figure 4.6 ^{14}C fixation
(Station A)


A
mg C m⁻³ hr⁻¹
60
40
20
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

programme.

ii. Station B:

Again the primary productivity fluctuations at this station followed the pattern obtained for chlorophyll a concentration seasonal variation (Fig. 4.7). A value of ca. 9 mg C m^{-3} hr^{-1} in September, 1982 was followed by a drop to a value of ca. 1 mg C m^{-3} hr^{-1}. The productivity remained at this level until April, 1983. The productivity increased in April and reached a maximum of ca. 41 mg C m^{-3} hr^{-1} in June. This high peak was followed by smaller peaks from July to November, 1983. During the winter of 1984, the low values of productivity remained at the same level of <1 mg m^{-3} hr^{-1} until the spring bloom which started in April. The productivity reached a maximum of ca. 46 mg C m^{-3} hr^{-1} in May, dropped to ca. 5 mg C m^{-3} hr^{-1} and increased again to ca. 22 mg C m^{-3} hr^{-1} in July. It dropped again and remained low during the following months.

IV. Total Number of Zooplankton

i. Station A:

Throughout the 28 months of sampling, the number of zooplankton changed from time to time. In this study, the number of zooplankton exceeded 500 cell m^{-3} on one occasion only (Fig. 4.8). From September, 1982 to March, 1983, the total number varied from 0 to ca. 100 cell m^{-3}. This figure increased in April, 1983 when it reached ca. 250 cell m^{-3}. The maximum number of organisms was found in May, 1984, when it reached ca. 2300 cell m^{-3}. Then it decreased throughout the winter months with the exception of one peak in October, 1983. In the spring of 1984 there was a slight increase to more than 100 cell m^{-3} in March and April. By

Figure 4.7 ^{14}C fixation
(Station B)

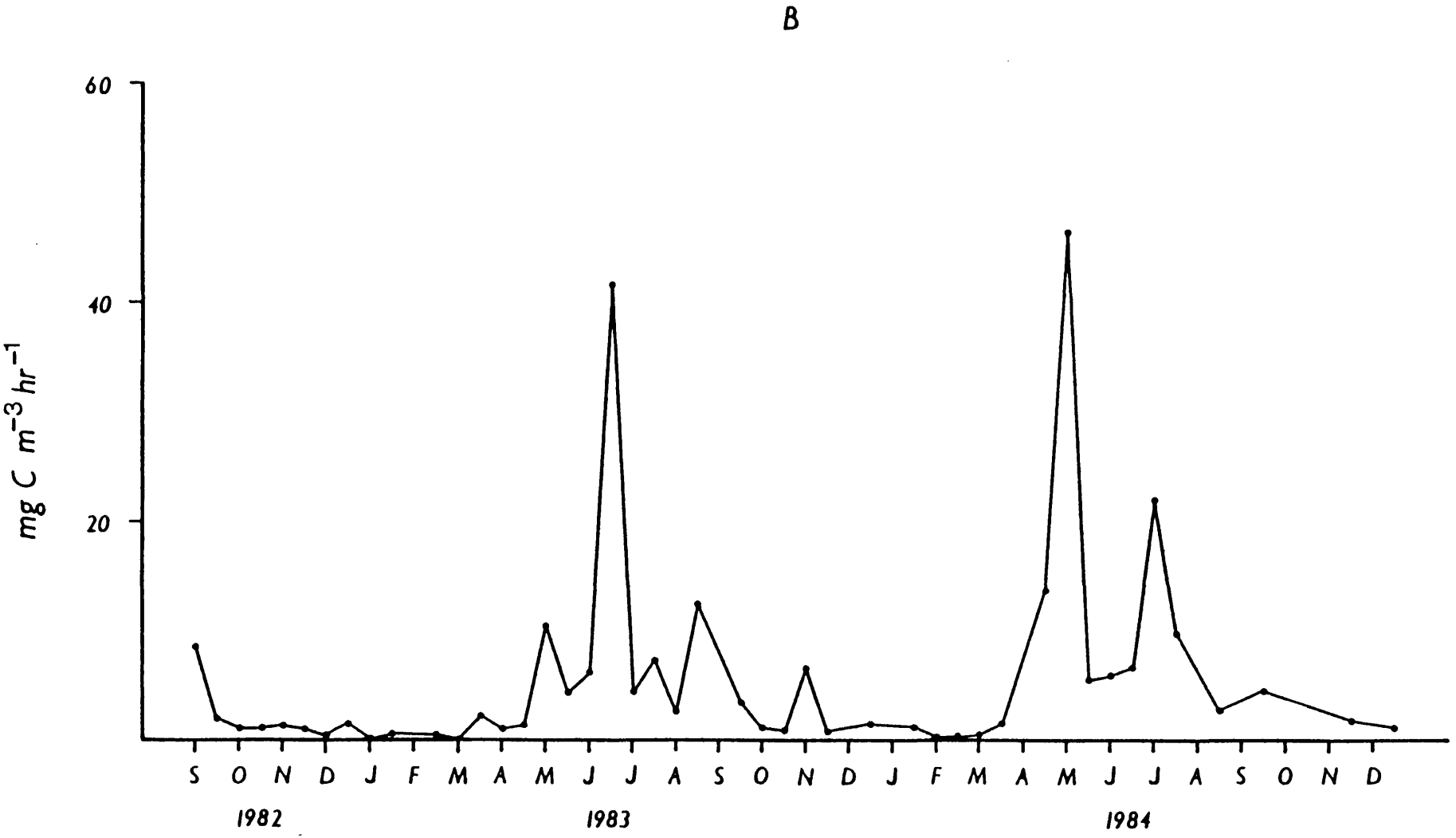
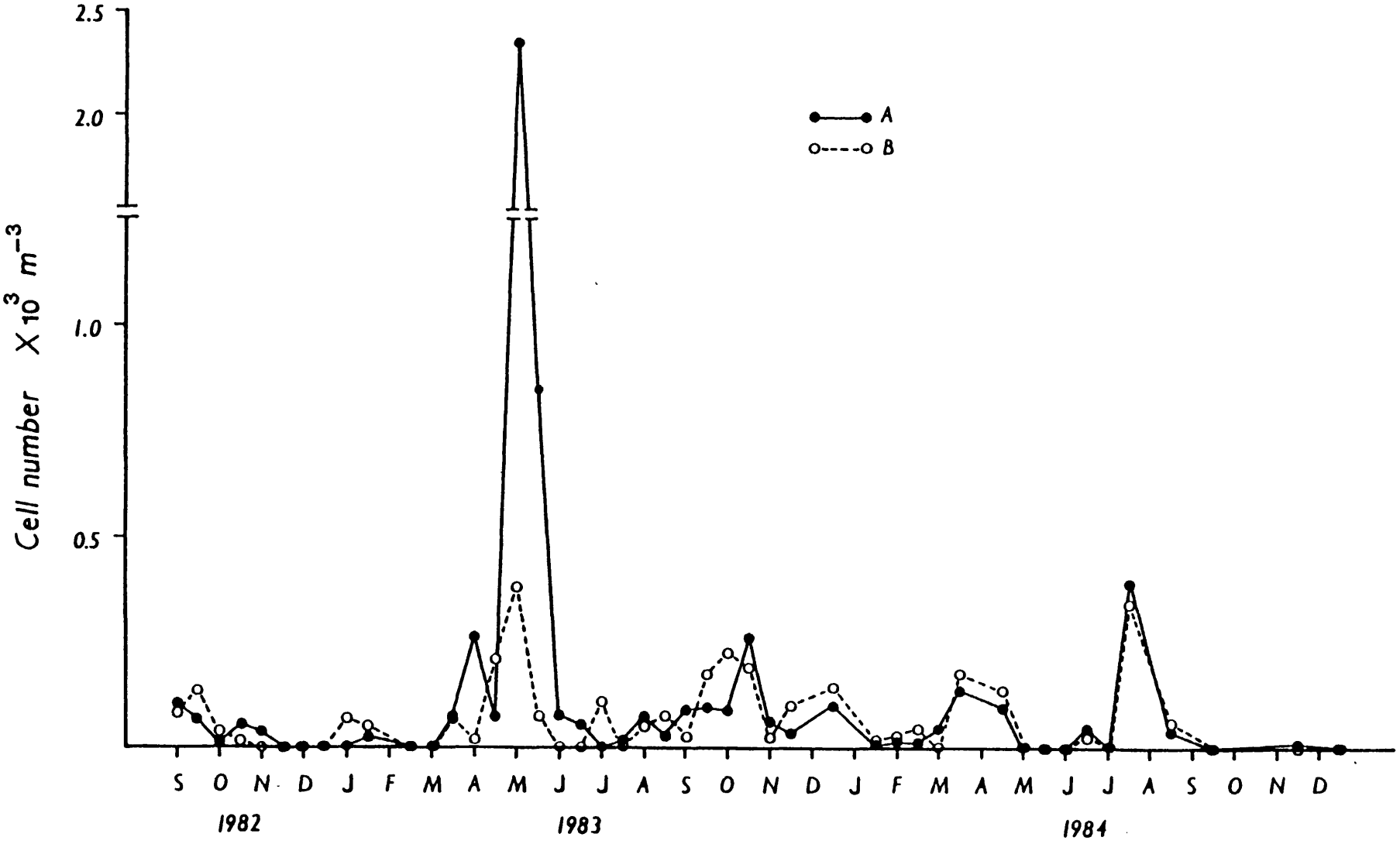

B
60
40
20
mg C m^{-3} hr^{-1}
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

Figure 4.8 Zooplankton total cell number


2.5
2.0
1.0
0.5
Cell number X 10^3 m^-3
A
B
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

the end of the summer, a peak of ca. 400 cell m^{-3} was found in July. This peak was followed by very low values throughout the autumn and winter months of 1984.

ii. Station B:

The number of zooplankton in this station, throughout the period of this study, were lower than those found in Station A (Fig. 4.8). The low number of organisms which occurred from September, 1982 to March, 1983 were followed by a maximum number of ca. 400 cell m^{-3} in May. Low values of zooplankton cell number (ca. 50 cell m^{-3}) were found in June, July and August, 1983. These were followed by an increase to ca. 200 cell m^{-3} in September and October, and to ca. 150 cell m^{-3} in December, 1983. The cold months of 1984 winter were followed by a slight increase in the number of zooplankton to ca. 150 cell m^{-3} in March. The highest peak in 1984 was found later in July when the number reached ca. 400 cell m^{-3}.

DISCUSSION

I. Phytoplankton Numbers - Biomass

The results of Stations A and B represent the annual variations in the number of net plankton found in Swansea Bay during the period of sampling. In both stations diatoms dominated the populations of the phytoplankton.

The flagellates occurred during short periods of the year in relatively small numbers (Figs. 4.2 and 4.3). From the results represented in Figures 4.1, 4.4 and 4.5 it can be seen that on many

occasions, the total number of net plankton did not coincide with the biomass represented as mg chlorophyll *a* m^{-3}.

During the period from March to May, 1983, two high peaks of net plankton were found at the time when the biomass was low (Fig. 4.1, Station A).

The most obvious examples can be drawn from the samples collected in October, 1983 (A, B), March, 1984 (B) and June, 1984 (A) (Figs. 4.4 and 4.5).

These discrepancies in the results of phytoplankton numbers and phytoplankton biomass for both stations can be confirmed by the insignificant correlation for A ($r = +0.3875$) and B ($r = +0.384$) (Figs. 4.9 A and B)

In the study of inshore phytoplankton of Swansea Bay, Paulraj (1974) found that the total number of net plankton and concentrations of chlorophyll *a* were not correlated. It is widely accepted that nanoplankton are usually more abundant and productive than net plankton, especially in oceanic waters (Steemen Nielsen and Jensen, 1957; McAllister *et al.*, 1959; Malone, 1971b; Semina, 1972).

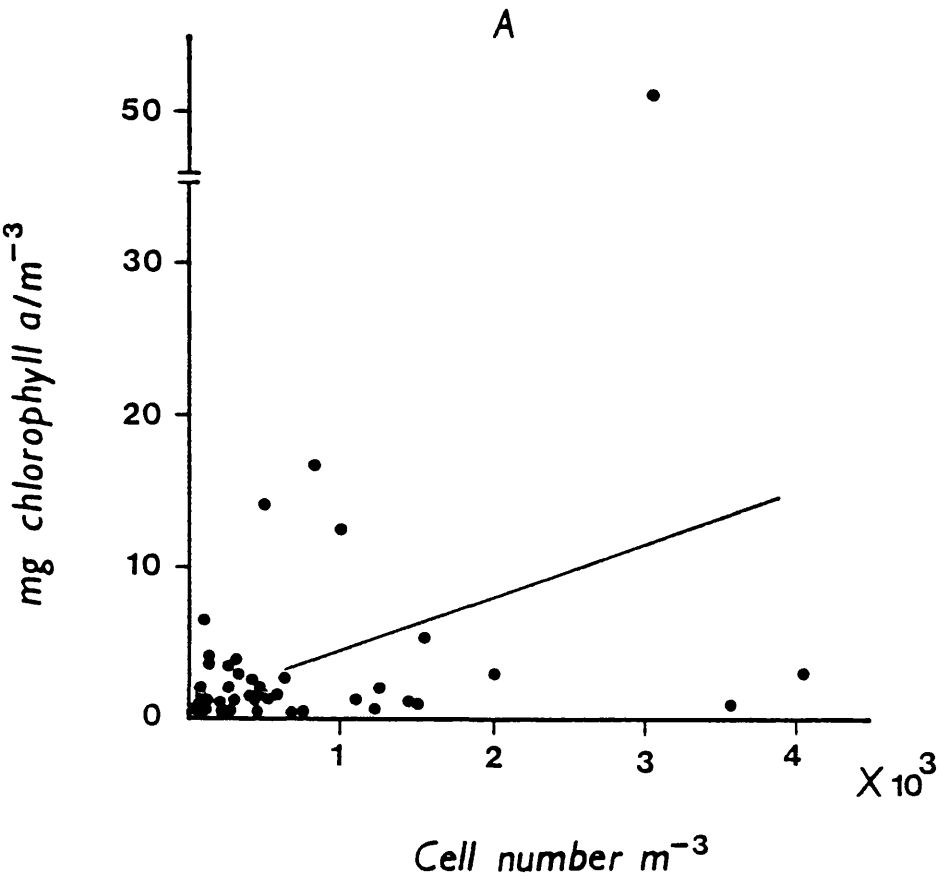
The net plankton dominate the continental shelf and coastal waters (Hasle, 1959; Hulbert, 1962,1970; Throndsen 1973). Exceptions to this general pattern have been observed in shallow, temperate estuaries and adjacent coastal waters influenced by estuarine runoff (Ryther, 1954; Loftus *et al.*, 1972; Durbin *et al.*, 1975; Malone, 1977b).

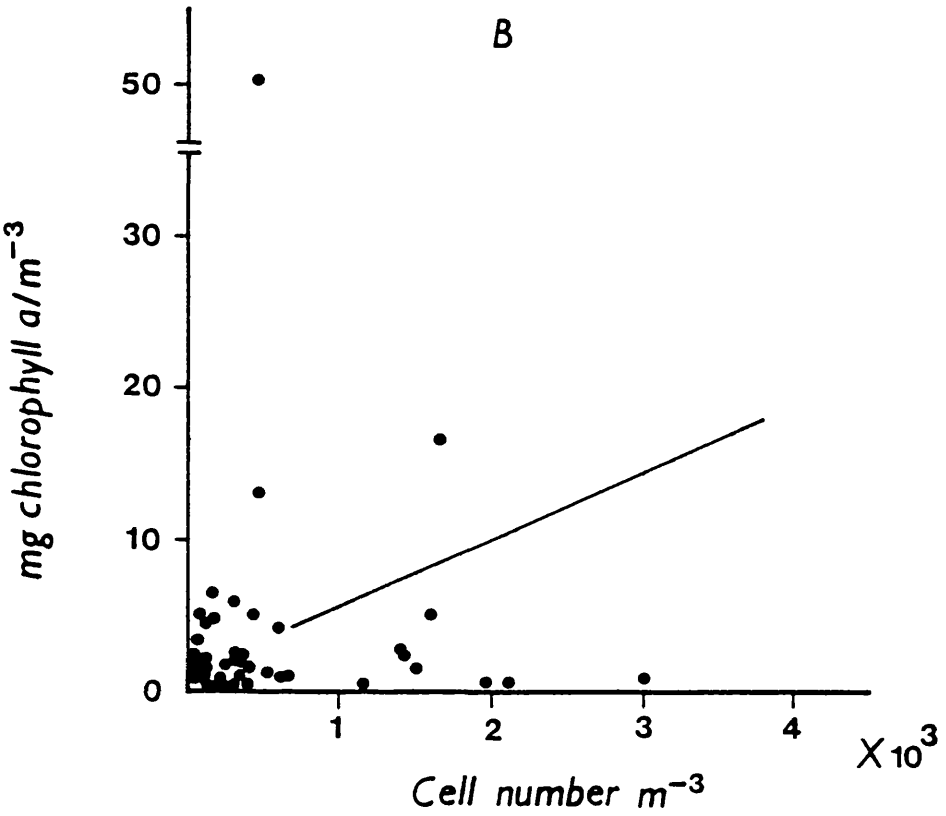
The disagreement between the results of phytoplankton cell number and phytoplankton biomass suggests that the latter situation mentioned above existed in Swansea Bay. This disagreement could be a result of the very small phytoplankton escaping from the net during sampling due to the relatively large mesh size (*ca.* 50 μm). Although some small

Figure 4.9 Correlation between net phytoplankton cell number and chlorophyll *a*

$r(A) = +0.39$

$r(B) = +0.38$


A
50
30
20
10
0
mg chlorophyll a/m^{-3}
1
2
3
4
X 10^3
Cell number m^{-3}


B
50
30
20
10
0
mg chlorophyll a/m^{-3}
1
2
3
4
X 10^3
Cell number m^{-3}

cells have been traced in the phytoplankton net collection, the total number of phytoplankton has been underestimated.

II. Phytoplankton Biomass - Primary Productivity

The results of this study show the annual variation in chlorophyll a concentration and primary productivity occurring in Swansea Bay. The values obtained for chlorophyll a are relatively high in comparison to those obtained by Paulraj (1974) who found values of 26.6 mg m^{-3} in an inshore area in Swansea Bay.

From 1973-1974, eleven cruises were carried out by I.M.E.R. in the Bristol Channel. The highest chlorophyll a content in the channel was found in Swansea Bay and around the Gower peninsula. The shallow area of Swansea and Carmarthen Bays provided the most suitable conditions for growth of phytoplankton where a value of 41 mg C m^{-2} hr^{-1} was found (I.M.E.R., 1974).

The values of primary productivity obtained in the present study (Figs. 4.6 and 4.7) are much higher than those found by Joint and Pomroy (1981) in the inner channel (3.3 mg C m^{-2} hr^{-1}), and the central channel (28.1 mg C m^{-2} hr^{-1}).

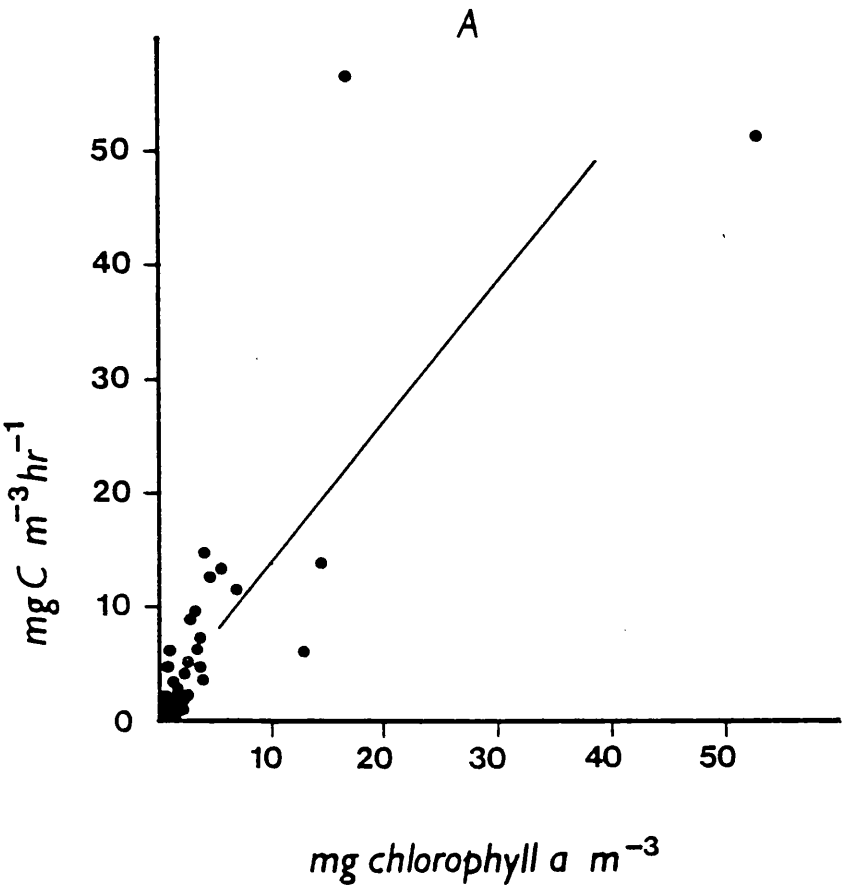
In general, the values of total primary production for both stations were the same. On a few occasions, differences were found which could be a result of the spatial heterogeneity in the bay.

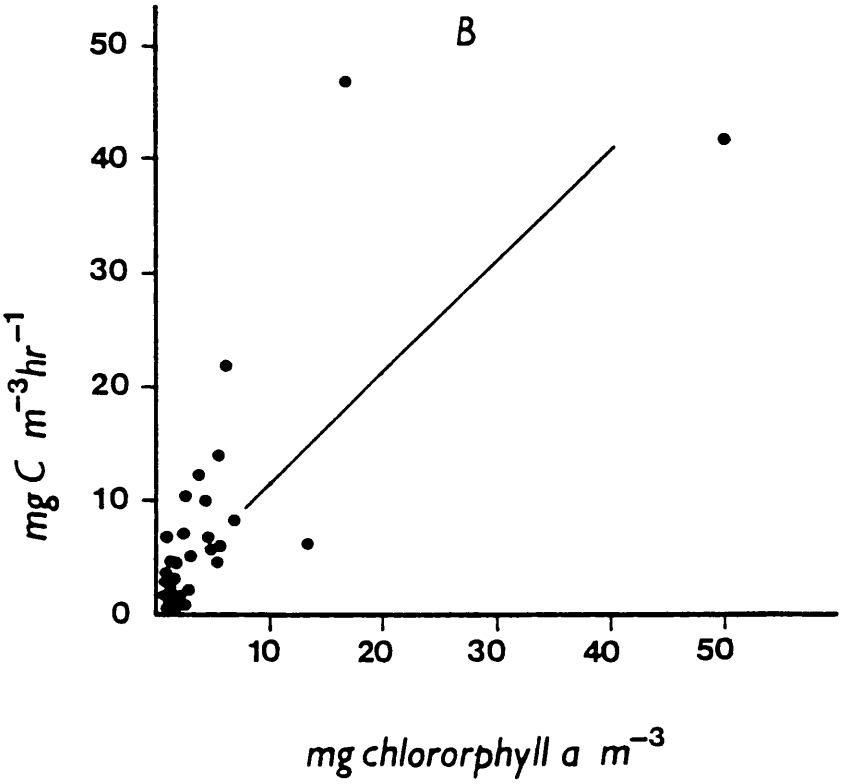
The relation between biomass and primary productivity exhibited a significant positive correlation (rA = +0.822, rB = +0.7921) (Figs. 4.10 A and B). Here we have a strong positive correlation between biomass and productivity. This emphasizes that the weak correlation obtained in both stations between cell number and chlorophyll a was a result of the underestimation of the actual number of phytoplankton in

Figure 4.10 Correlation between biomass and primary productivity

r(A) = +0.82

r(B) = +0.79


A
50
40
30
20
10
0
10
20
30
40
50
mg C m^{-3}hr^{-1}
mg chlorophyll a m^{-3}


B
50
40
30
20
10
0
10
20
30
40
50
mg C m^{-3}hr^{-1}
mg chlororphyll a m^{-3}

Swansea Bay.

The assimilation number (photosynthesis/biomass) of the phytoplankton populations during the period of sampling varied from 0.195 - 8.35 in Station A, and from 0.11 - 9.77 mg C mg chlorophyll *a* hr^{-1} (Table 4.2).

It has been demonstrated that temperature (Eppley, 1972), light intensity (Beardall and Morris, 1976) and nutrient limitation (Thomas and Dodson, 1972), directly affect phytoplankton assimilation numbers.

In the present study, relatively high assimilation numbers were obtained in November, 1982 (A), January, 1983 (A), September, 1983 (A), November, 1983 (B) and July, 1984 (A, B) coinciding with low numbers of net phytoplankton. Eppley (1972) proposed that high assimilation numbers were associated with cells of low cell volume and fast growth rates; and Malone (1971b) determined that nanoplankton from different oceanic areas consistently had higher assimilation numbers than net plankton. Based on the previous findings, it is possible to suggest that, when high assimilation numbers were obtained at the time when cell count of net phytoplankton was low, the nanoplankton may have played an important role in the productivity of Swansea Bay.

III. Phytoplankton- Zooplankton

One of the factors which could either by itself or in association with the other environmental factors affect phytoplankton biomass, is zooplankton. The effect of zooplankton as grazers had been identified as early as 1935 (Harvey *et al.*, 1935; Riley, 1946).

The zooplankton bloom which took place in May 1983 at Station A was mainly dominated by copepods. This bloom with an abundance of *ca.* 2300 cell m^{-3} was the highest found during this study (Fig. 4.8). This

Table 4.2 Assimilation number (productivity/biomass)

DATE	A	B	DATE	A	B
7. 9.82	0.97	1.29	22. 9.83	7.32	5.23
22. 9.82	0.84	0.85	10.10.83	1.37	0.7
5.10.82	1.23	0.42	19.10.83	3.74	1.22
19.10.82	1.13	0.49	4.11.83	8.35	9.77
2.11.82	4.1	0.95	18.11.83	1.47	1.87
16.11.82	1.11	0.55	12.12.83	0.7	2.46
3.12.82	0.42	0.27	24. 1.84	0.3	0.87
14.12.82	0.58	0.92	3. 2.84	0.53	0.22
25. 1.83	4.37	0.46	17. 2.84	3.26	1.08
7. 2.83	3.57	1.05	20. 3.84	1.22	2.28
31. 3.83	0.92	2.13	30. 4.84	2.55	2.69
12. 4.83	0.66	0.83	14. 5.84	3.37	2.78
26. 4.83	1.0	0.9	31. 5.84	1.78	1.27
11. 5.83	3.12	4.26	13. 6.84	1.33	1.15
25. 5.83	1.6	2.65	27. 6.84	2.2	1.56
10. 6.83	0.47	0.48	11. 7.84	3.39	3.66
23. 6.83	0.98	0.82	30. 7.84	3.74	2.52
11. 7.83	2.94	1.66	30. 8.84	2.06	2.55
26. 7.83	2.36	3.57	17. 9.84	2.04	2.39
22. 8.83	1.48	4.0	13.11.84	1.68	1.9
30. 8.83	2.08	3.54	11.12.84	1.43	1.13

peak occurred after a period of low productivity and low biomass (chlorophyll *a*).

On the other hand this bloom was preceded in April by a relatively high number of net phytoplankton cells (1500 cell m^{-3}). Since macro-zooplankton selectively remove large particles from multi-sized algal diets (Mullin, 1963; Frost, 1972; Gaudy, 1974) it can be said that the decline of the net phytoplankton in the sample directly preceding the zooplankton bloom was due to zooplankton selectively grazing the larger cells (Chervin, 1978). Plankton cell numbers in Station B showed a similar pattern of phytoplankton and zooplankton alternative domination but to a lesser extent. The highest zooplankton peaks of 1984 at both stations were of similar values and occurred in July following a bloom dominated by *Rhizosolenia delicaltula* (95%, A; 71%, B). The peak at Station A was not as high as the one which took place in May, 1983 following a bloom of *Rhizosolenia hebetata* (48%), and *Biddulphia sinensis* (28%). The peak of zooplankton at Station B in 1984 was as high as the peak of 1983 which followed a bloom of *Biddulphia sinensis* (22%) and *Coscinodiscus sp.* (22%). From the results (Figs. 4.1 and 4.8), it seems that zooplankton in Swansea Bay were selective grazers and prefer relatively large phytoplankton cells. This can be seen from the high peak of zooplankton (Station A) which followed a phytoplankton bloom in 1983 dominated by large phytoplankton cells (*R. hebetata*, 48%). The presence of *B. sinensis* in numbers (22%) suitable to support a growth of zooplankton in 1983 resulted in the occurrence of medium zooplankton peak (May 1983). On the other hand, the onset of *R. delicatula* bloom in June 1984 (*ca.* 80%) did not support the increase of zooplankton numbers to a high level as the one achieved in May, 1983 (A).

CHAPTER V

PHYTOPLANKTON SPECIES SUCCESSION

INTRODUCTION

Succession is a process of continuous changes occurring in the ecosystem. In the sea, the changes occur at different levels. The taxonomic composition of phytoplankton communities, and the abundance and relative dominance of the different species and algal groups present undergo continuous change. Diatoms usually dominate in colder, nutrient-rich waters. The dominance of diatoms is sometimes replaced by dinoflagellates and coccolithophorids. This succession of phytoplankton groups often occurs in regions characterized by seasonal changes in temperature and nutrients (Gran and Braarud, 1935).

In addition to phytoplankton group succession, species succession plays a very important and fundamental role in the ecology of phytoplankton. Throughout the species succession, few or many species may occur together. One or a group of species may dominate the community. The duration and time of occurrence and disappearance of different species vary.

In temperate regions the annual variation of phytoplankton biomass and growth follow a defined pattern. In the winter, there is no appreciable growth. Phytoplankton numbers increase early in the spring reaching a maximum towards the end of April. The peak is usually followed by a sharp decline, and during the summer numbers remain at a relatively low level. A second maximum, usually not as high as the spring maximum, may occur in the autumn, after which the numbers decrease to the low winter level.

The timing and magnitude of the blooms occurring in the temperate region depend on a number of environmental factors. The individual or collective effect of the different environmental factors on the growth

and succession of phytoplankton species is specific for each geographical region. Some of the most important factors are:

a. Temperature and light:

Temperature and light affect phytoplankton species differently. Each species has its own requirement of temperature and light. In addition, the different combinations of temperature and light stimulate different responses. The combinations of temperature and light vary according to season: in spring, an increase in light and low temperature; summer, an increase in light and high temperature; autumn, a decrease in light and high temperature; winter, a decrease in light and low temperature (Strickland, 1965; Hutchinson, 1967; Fogg, 1975).

b. Concentration of nutrients:

Nutrients have been known for a long time as one of the most important ecological factors that could affect phytoplankton growth. In addition to their role as growth-limiting factors, the availability of one nutrient at the expense of another may promote the growth of a specific group of phytoplankton organisms. For example, the bloom of diatoms usually exhausts the silica available for growth. The resulting new proportion of silica to other nutrients may support the growth of another group of phytoplankton such as Chlorophyceae (Fogg, 1975).

c. Hydrographical conditions:

The turbulence which results from the autumn and winter gales disturbs the stable water column which developed in the summer. It is possible that the turbulent conditions favour rapidly sinking forms and the stabilized water column favours buoyant and motile algae (Moss, 1969).

In addition to the effect of turbulence on buoyancy, these conditions can result in increased circulation of nutrients in the water mass.

d. Selective grazing:

The biomass and species composition of phytoplankton are influenced largely by the presence of grazers in the water column. Grazers may be selective or non-selective (Edmondson, 1964). During the period of high growth rate of grazers, and when conditions favour selective grazing, phytoplankton species of a certain cell size may be replaced by a differently sized species (Frost, 1977).

Phytoplankton species succession has been studied throughout the Bristol Channel (Rees, 1939; Pearce, 1967; Paulraj, 1974; I.M.E.R., 1975; Tyler, 1976; Vogelmann, 1980; Sexton, 1985). The phytoplankton of inshore Swansea Bay have been studied extensively (Paulraj, 1974) and were found to exhibit a regular annual pattern of species succession.

In the present study, ecologically significant species (those species which made up at least 30% of a phytoplankton net sample, Vogelmann (1980)) were related to the environmental factors. This was carried out to see if there was a correspondence between the dominant phytoplankton species and the environmental conditions existing at the time of sampling.

RESULTS

Phytoplankton abundance, temporal variation and species succession are essential topics in the study of aquatic primary productivity.

In the present study, the data of phytoplankton abundance throughout the period of study were treated in two ways. Firstly, the total number of each species in each sample was represented as a percentage of the highest total number of phytoplankters which occurred during the period of study. This will give an idea of the relative importance of each species and its temporal variations. Secondly, the bimonthly samples were treated as sub-units. The total number of each species in each sample was represented as a percentage of the total number of phytoplankton in that sample. This method will show the dominant species in each sample.

During the study period, 43 phytoplankton species were identified. Any species representing 30% or more of the population at least once during the study period was considered dominant. Twelve species have fallen into this category.

1. *Bacillaria paxillifer*

At both stations, the occurrence of this species took place during the spring and autumn seasons. An exception was the autumn of 1984.

Station A (Figures 5.1.1 and 5.1.2)

Figure 5.1.1 represents the temporal variation of *B. paxillifer*. Throughout the period of sampling, five peaks of this species occurred at Station A. The first one occurred in October, 1982 (6.5%); followed by May, 1983 (21%); October, 1983 (11%); January, 1984 (3%); and March-

Figure 5.1.1 Temporal variation of Bacillaria paxillifer (Station A)

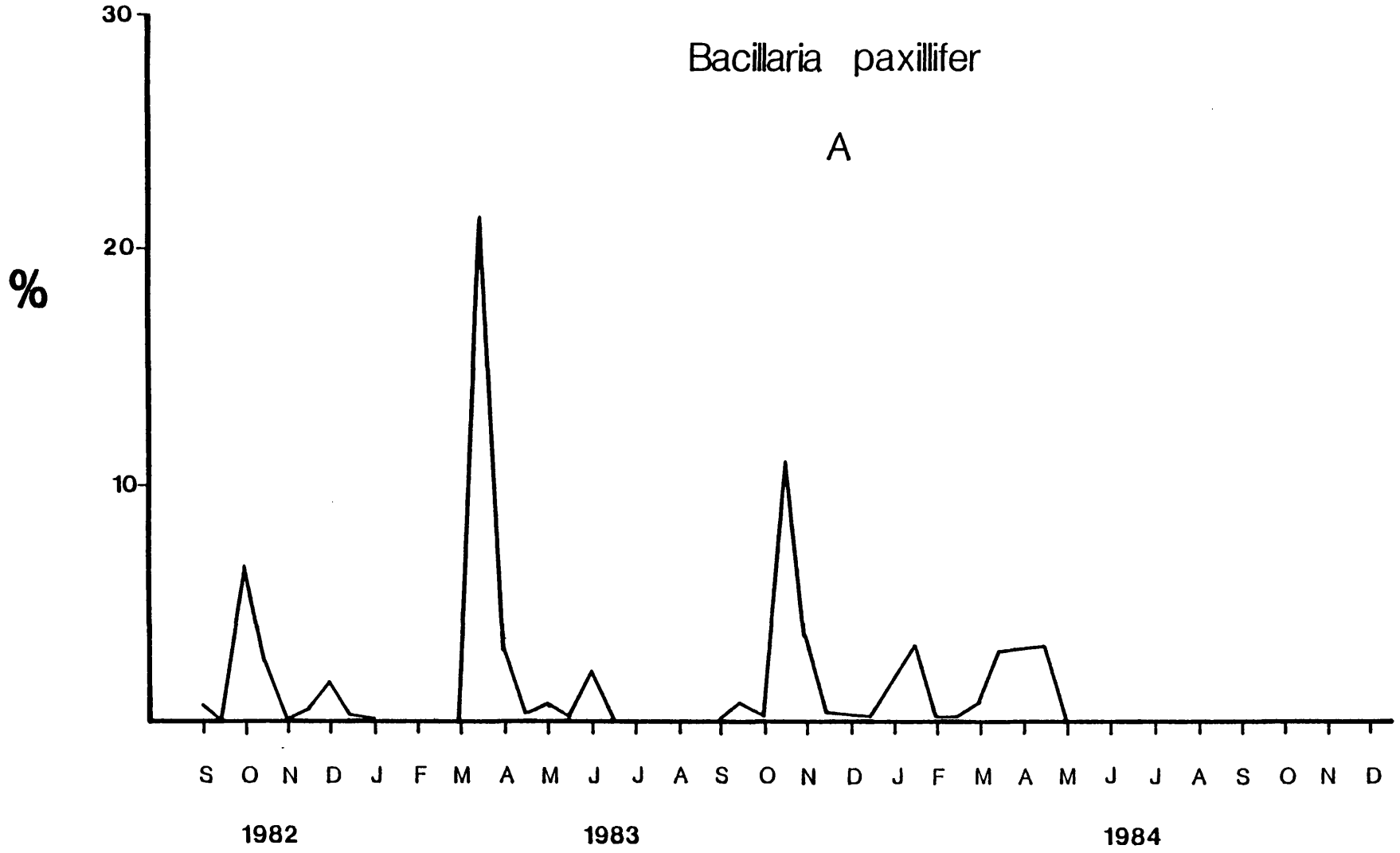
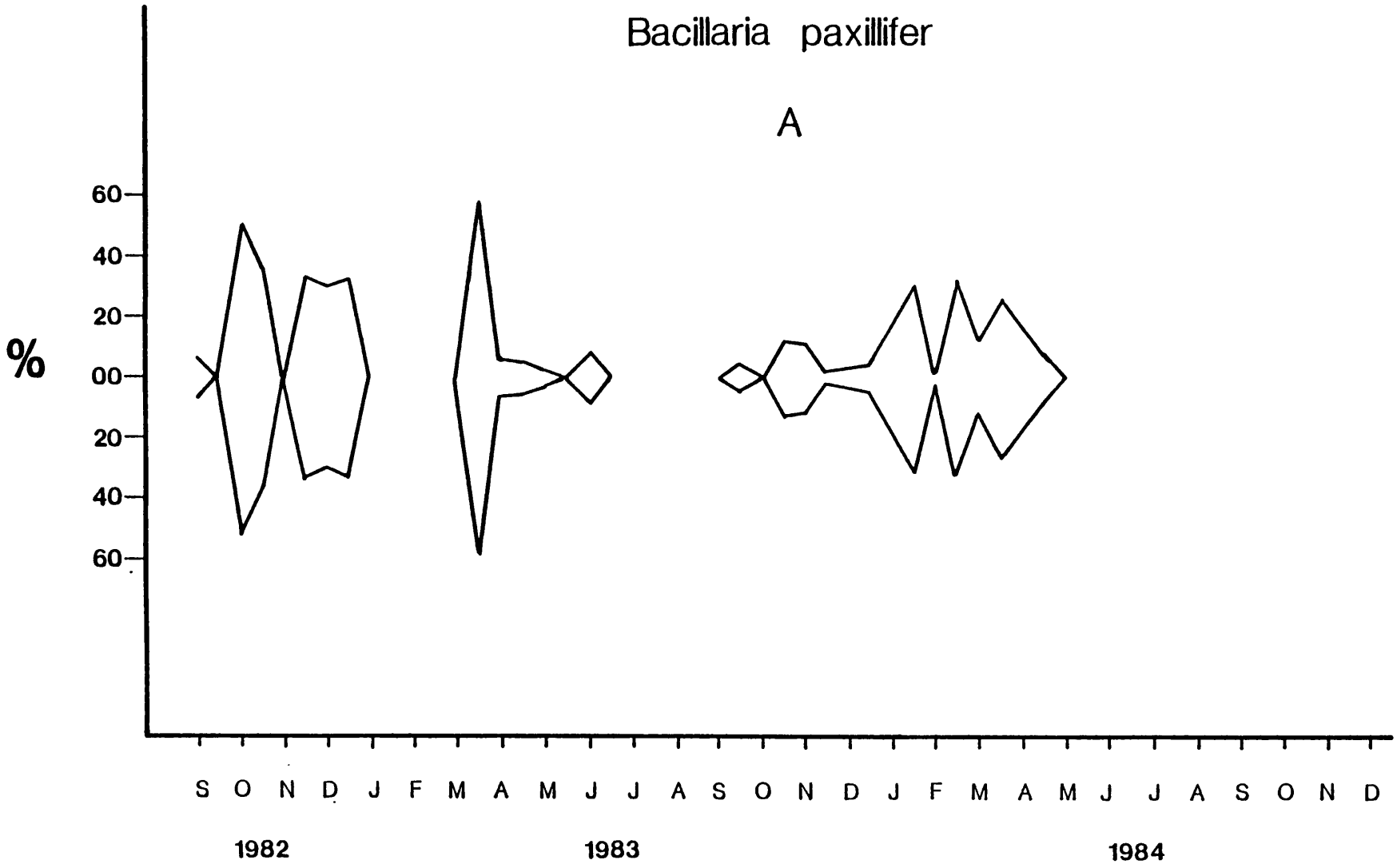

Bacillaria paxillifer
A
%
30
20
10
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

Figure 5.1.2 Dominance of Bacillaria paxillifer
(Station A)


Bacillaria paxillifer
A
%
60
40
20
00
20
40
60
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

April, 1984 (3%).

B. paxillifer dominated the phytoplankton population at Station A in October, 1982 (50%); May, 1983 (56%), January and March, 1984 (32%). See Figure 5.1.2.

From Figures 5.1.1 and 5.1.2 it can be seen that when the temporal variation was 2% in December, 1982, B. paxillifer dominated and represented 32% of the population.

Station B (Figures 5.1.3. and 5.1.4)

Figure 5.1.3 represents the temporal variation of B. paxillifer at Station B. It occurred from September, 1982 until July, 1983, and from September, 1983 until May, 1984. Some significant peaks took place during those periods; December, 1982 (3.5%); July, 1983 (16%); October, 1983 (9%); January, 1984 (6%); and March, 1984 (25%).

This species dominated the population in October, 1982 (36%); November, 1982 (50%); January, 1983 (30%); July, 1983 (45%); January, 1984 (60%) and March, 1984 (50%). See Figure 5.1.4.

By comparing Figures 5.1.3 and 5.1.4 it can be seen that in Figure 5.1.4 the dominance values of November, 1982 (50%), July, 1983 (45%) and March, 1984 (50%) correspond to temporal variation values of 0.5%, 16% and 25% respectively (Fig. 5.1.3).

2. Biddulphia sinensis

At both stations, this species occurred throughout the period of sampling from September, 1982 until June, 1984. It disappeared during the period July to December, 1984.

Figure 5.1.3 Temporal variation of Bacillaria paxillifer (Station B)

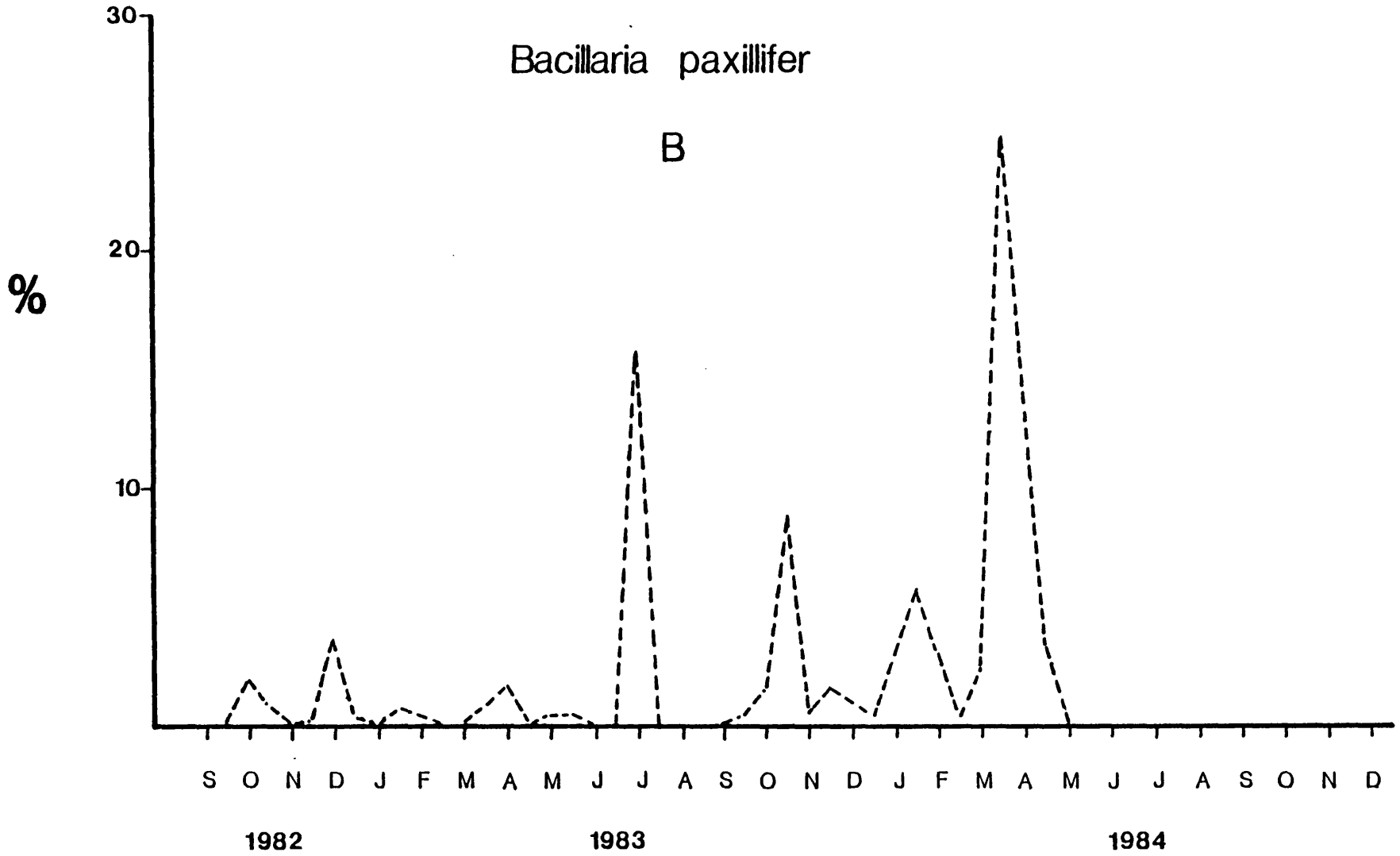
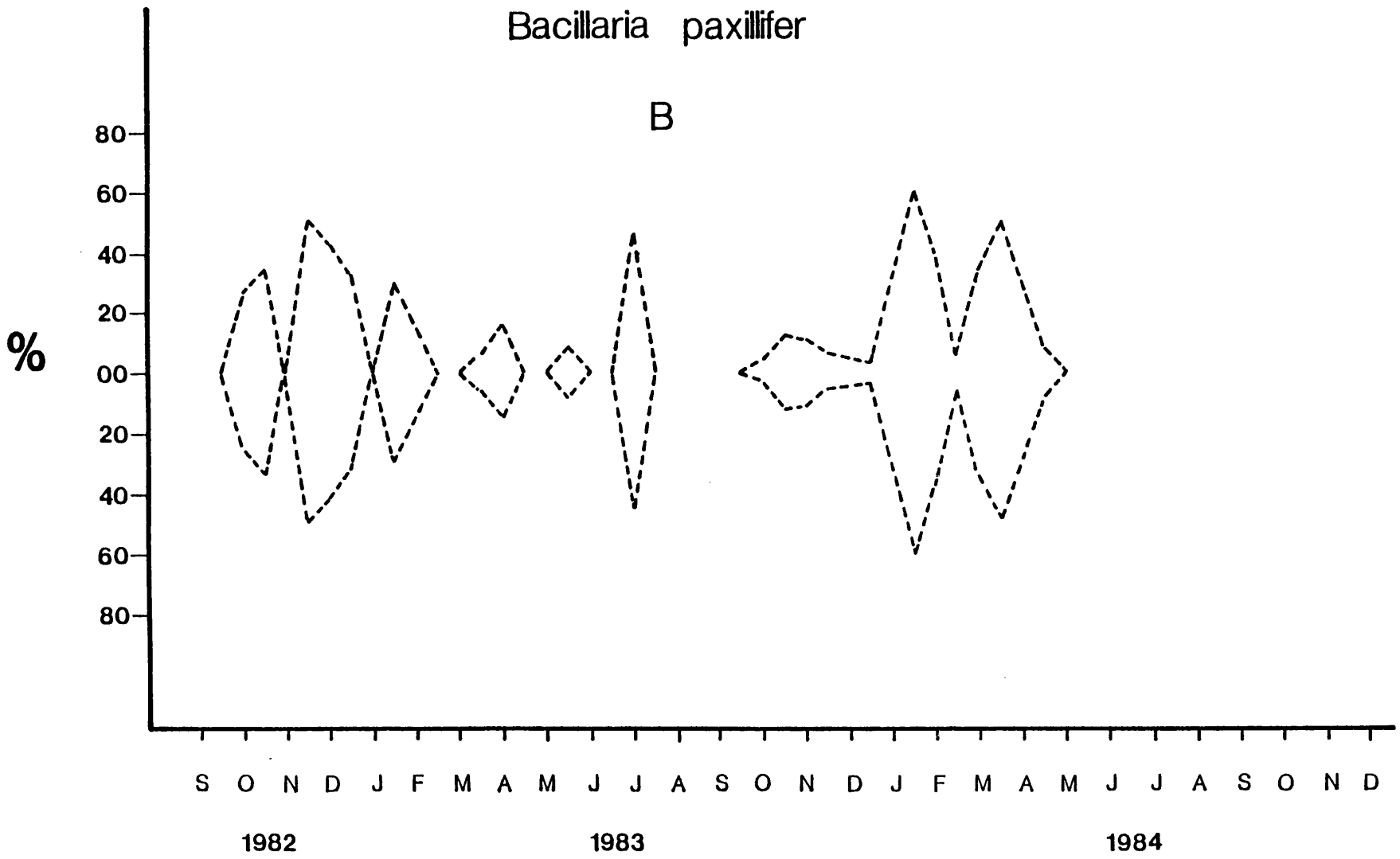

Bacillaria paxillifer
B
%
30
20
10
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

Figure 5.1.4 Dominance of Bacillaria paxillifer
(Station B)


Bacillaria paxillifer
B
%
80
60
40
20
00
20
40
60
80
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

Station A (Figures 5.2.1 and 5.2.2)

The temporal variation of B. sinensis at this station ranges from 0% to 72%. The most significant peaks occurred in September, 1982 (7%); November, 1982 (5.5%); April, 1983 (10%); May, 1983 (9%); October, 1983 (72%); January, 1984 (5.5%); and March, 1984 (4%). See Figure 5.2.1.

Biddulphia sinensis dominated the phytoplankton population at this station from September, 1982 until December, 1982 (43-60%). From January, 1983 until March, 1983 (75-98%); May, 1983 (34%) and September 1983 (41%). It dominated the population from October, 1982 until January, 1984 ranging from 50-94% of the population. From February, 1984 until March, 1984 it ranged from 30-66% of the population. See Figure 5.2.2.

Station B (Figures 5.2.3 and 5.2.4)

The values of temporal variation at this station were lower than those of Station A from September, 1982 until July, 1983. The maximum value in this period was 4% in October. The variation from September, 1983 until May 1984 took a very different pattern. The values were much higher than those in the first period. A peak of ca. 60% occurred in October, followed by a peak of 26% in November and a third one of ca. 10% in May.

A comparison of Figures 5.2.3 and 5.2.4 shows that although the values of temporal variations were relatively low in the first period (Fig. 5.2.3), the corresponding values in Figure 5.2.4 were very high. They reached 58% in October, 1982; 49% in November, 1982; 67% in December, 1982; 60% in February, 1983 and 32% in May, 1983. On the other hand, the period from September, 1983 until May, 1984 reflects a relatively normal correspondence. The values of 60%, 4%, 26% and 10%

Figure 5.2.1 Temporal variation of Biddulphia sinensis
(Station A)

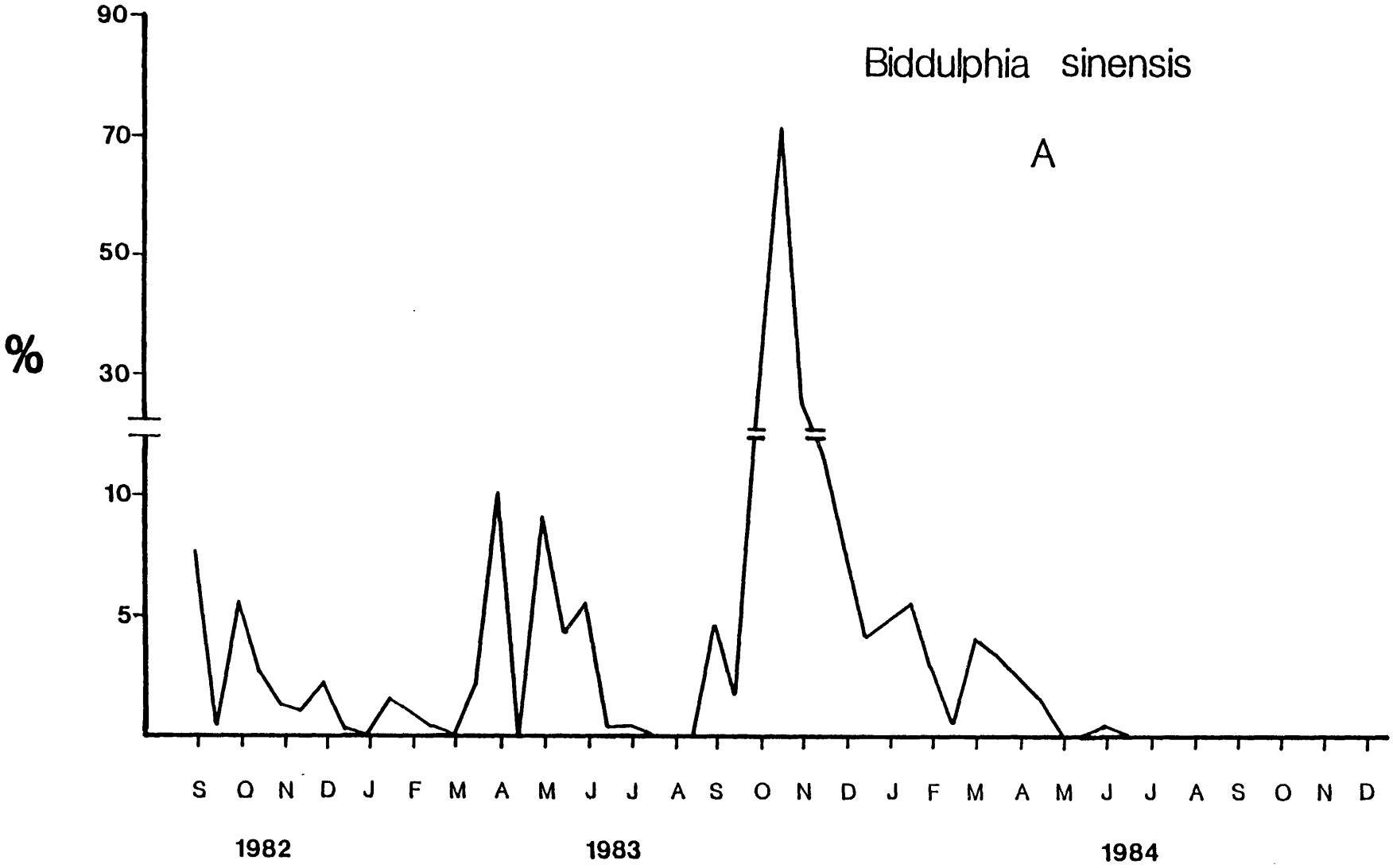

Biddulphia sinensis
A
%
90
70
50
30
10
5
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

Figure 5.2.2 Dominance of Biddulphia sinensis
(Station A)

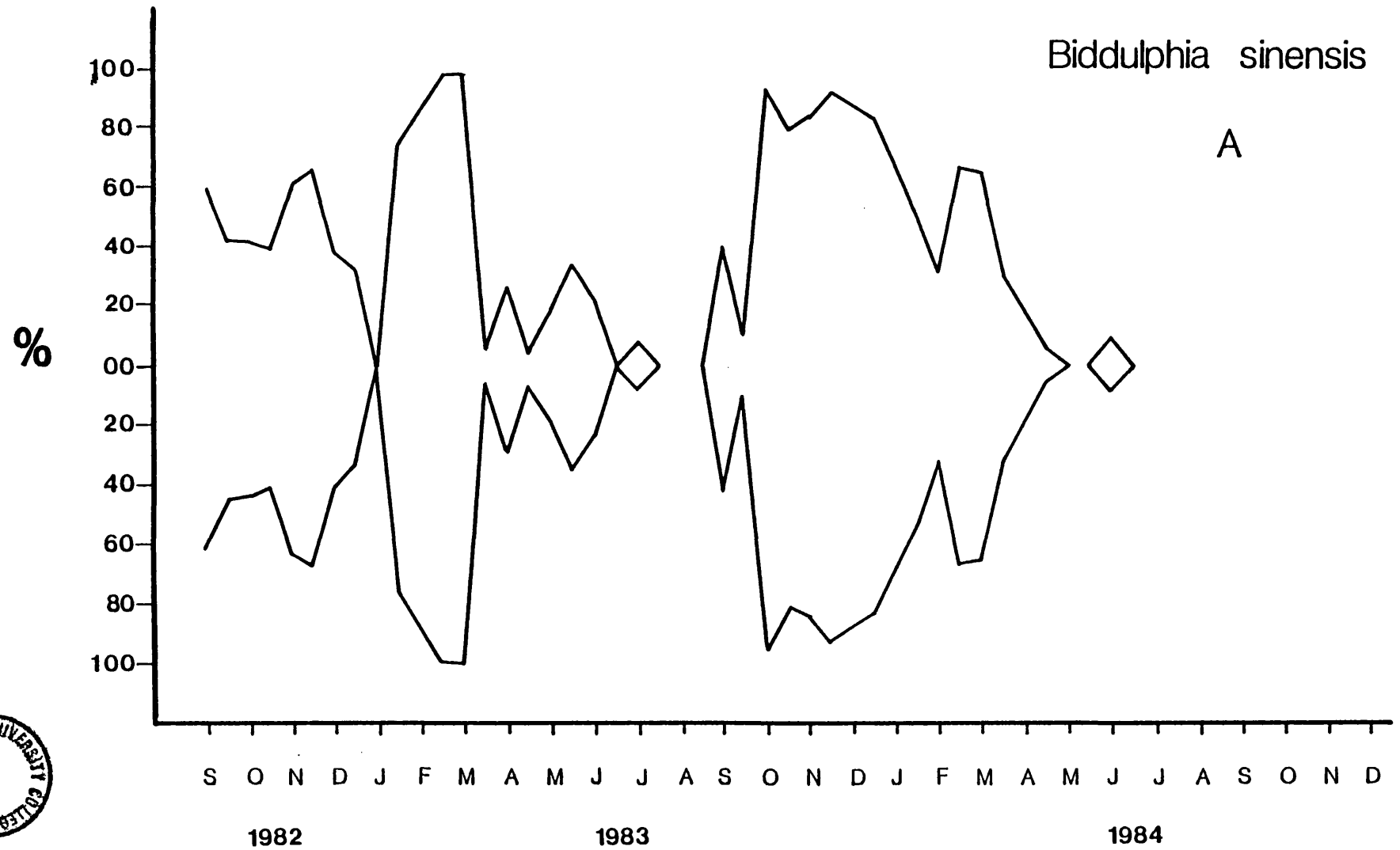


Figure 5.2.3 Temporal variation of Biddulphia sinensis
(Station B)

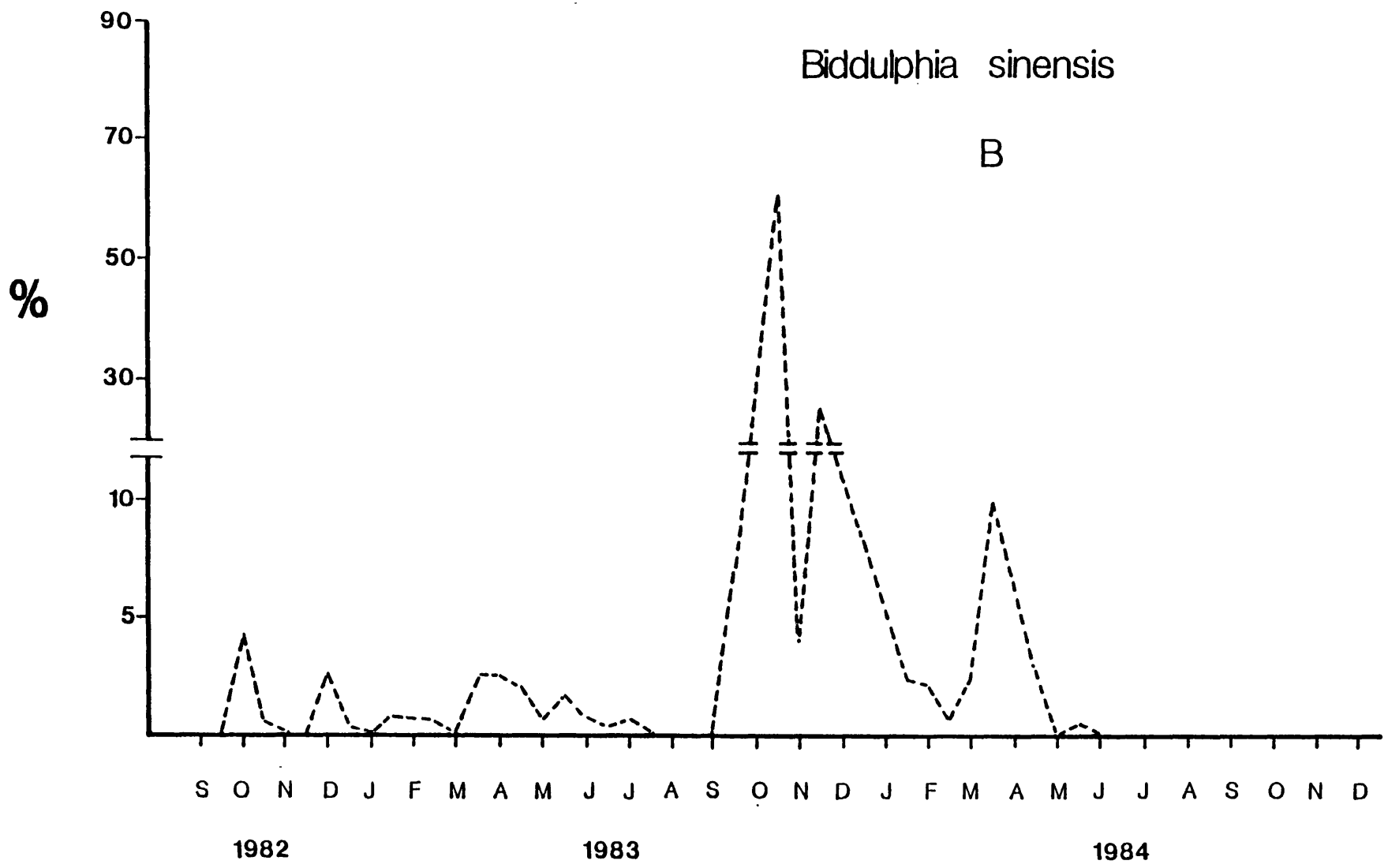
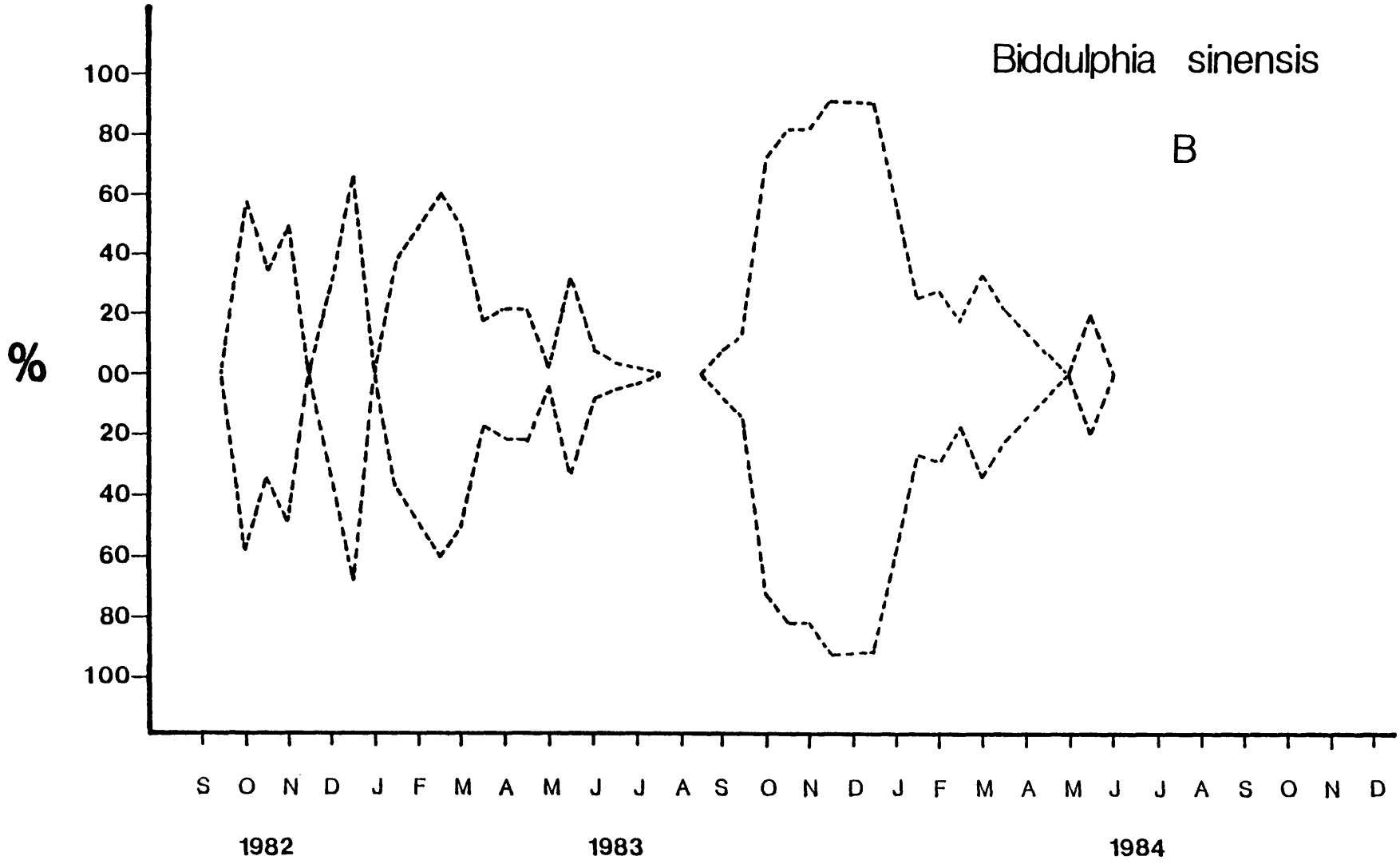

Biddulphia sinensis
B
%
90
70
50
30
10
5
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

Figure 5.2.4 Dominance of Biddulphia sinensis
(Station B)


Biddulphia sinensis
B
%
100
80
60
40
20
00
20
40
60
80
100
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

in Figure 5.2.3 corresponded to 73% (October, 1983); 82% (November, 1983); 92% (November, 1983) and 22% in March, 1984. The obvious exception in this period was the first value in November, 1983.

3. Coscinodiscus sp.

Coscinodiscus sp. occurred mainly in the spring and autumn seasons during the sampling period.

Station A (Figures 5.3.1 and 5.3.2)

At this station, the temporal variation of Coscinodiscus sp. was characterized by higher values in the spring than those found in the autumn. The values of spring 1983 reached a value of 4% in May, 1983; 5.5% in June, 1983; and 7% in April, 1984. On the other hand, the values of the autumn rarely exceeded 1%. Although the total number of Coscinodiscus sp. was low in January, 1983 (< 0.2%) and May, 1984 (0.5%), it dominated the sparse phytoplankton in those months, reaching 50% and 34% respectively.

Station B (Figures 5.3.1 and 5.3.3)

At this station, the temporal variation of Coscinodiscus sp. was similar to that of Station A with the exception of the September, 1983 value. At this station the temporal variation (Fig. 5.3.1) was characterized by low values of the autumn not reaching 1% except in September, 1983 when it reached 37.5%. The spring values reached 4.5% in May, 1983 and 9% in April, 1984.

Coscinodiscus sp. dominated the phytoplankton population in September, 1982 (65%); March, 1983 (50%); May, 1983 (33%) and September, 1983 (72%). From Figures 5.3.1 and 5.3.3, it can be seen

Figure 5.3.1 Temporal variation of Coscinodiscus sp.
(Stations A and B)

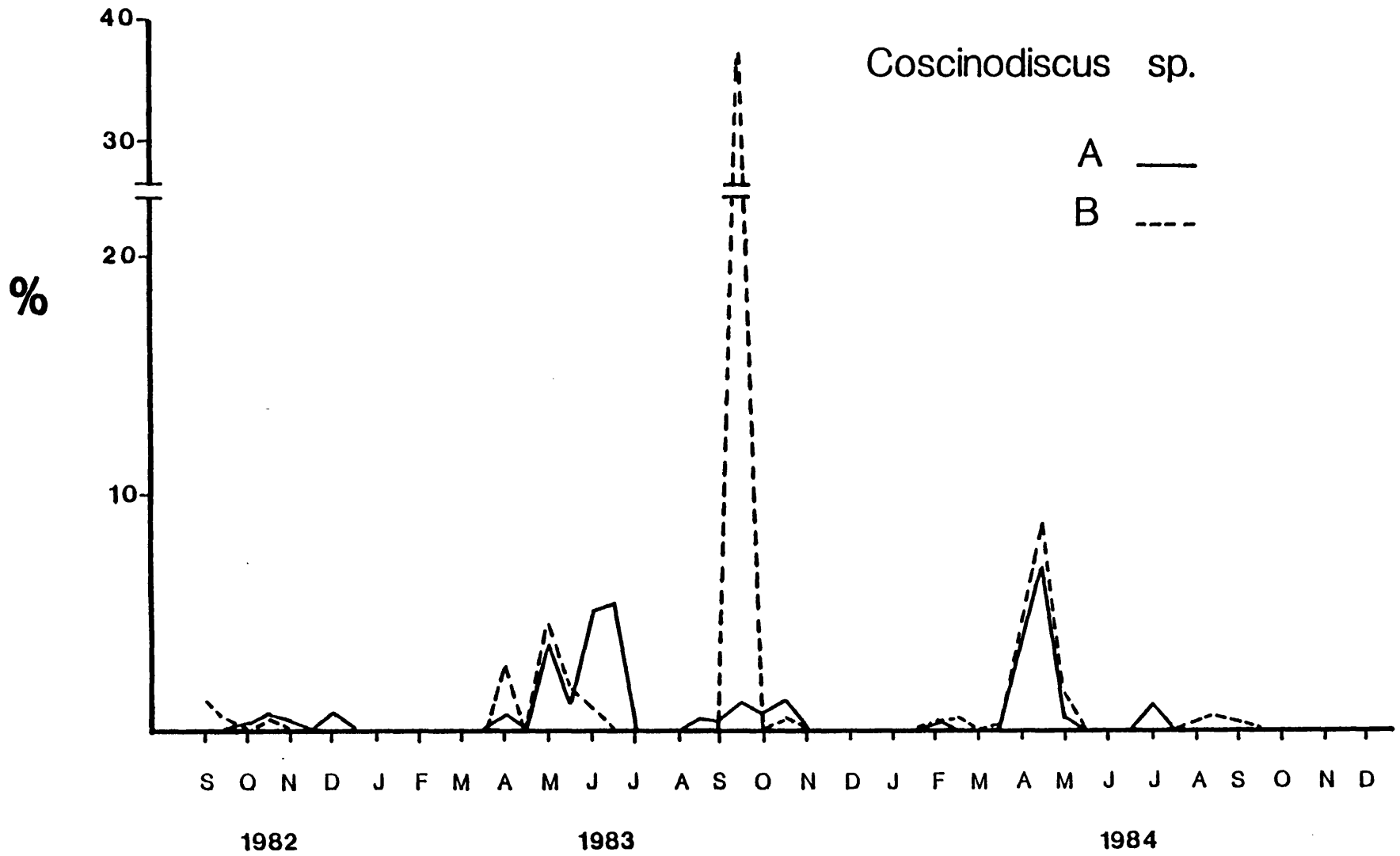

Coscinodiscus sp.
A
B
%
40
30
20
10
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

Figure 5.3.2 Dominance of Coscinodiscus sp.
(Station A)

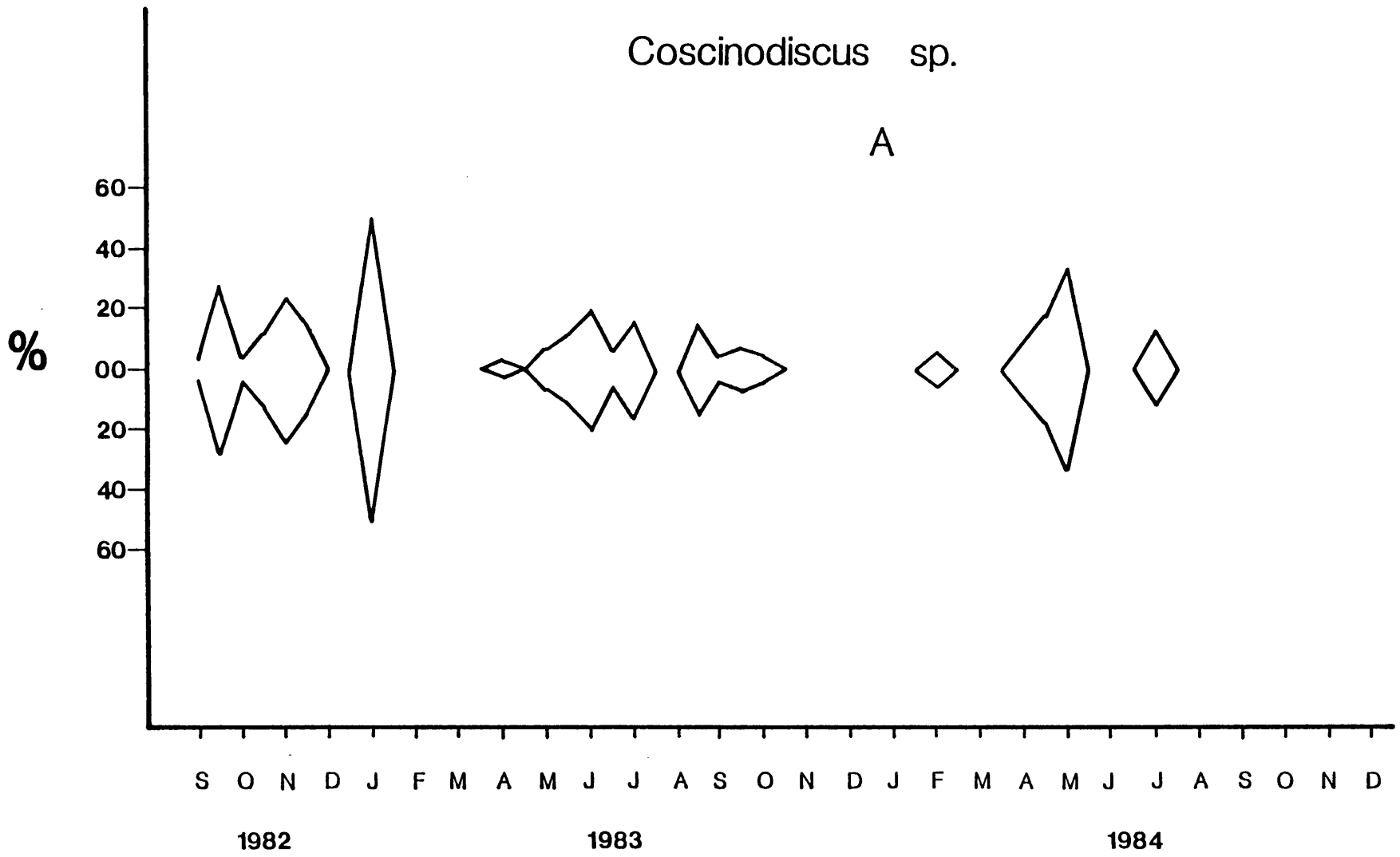
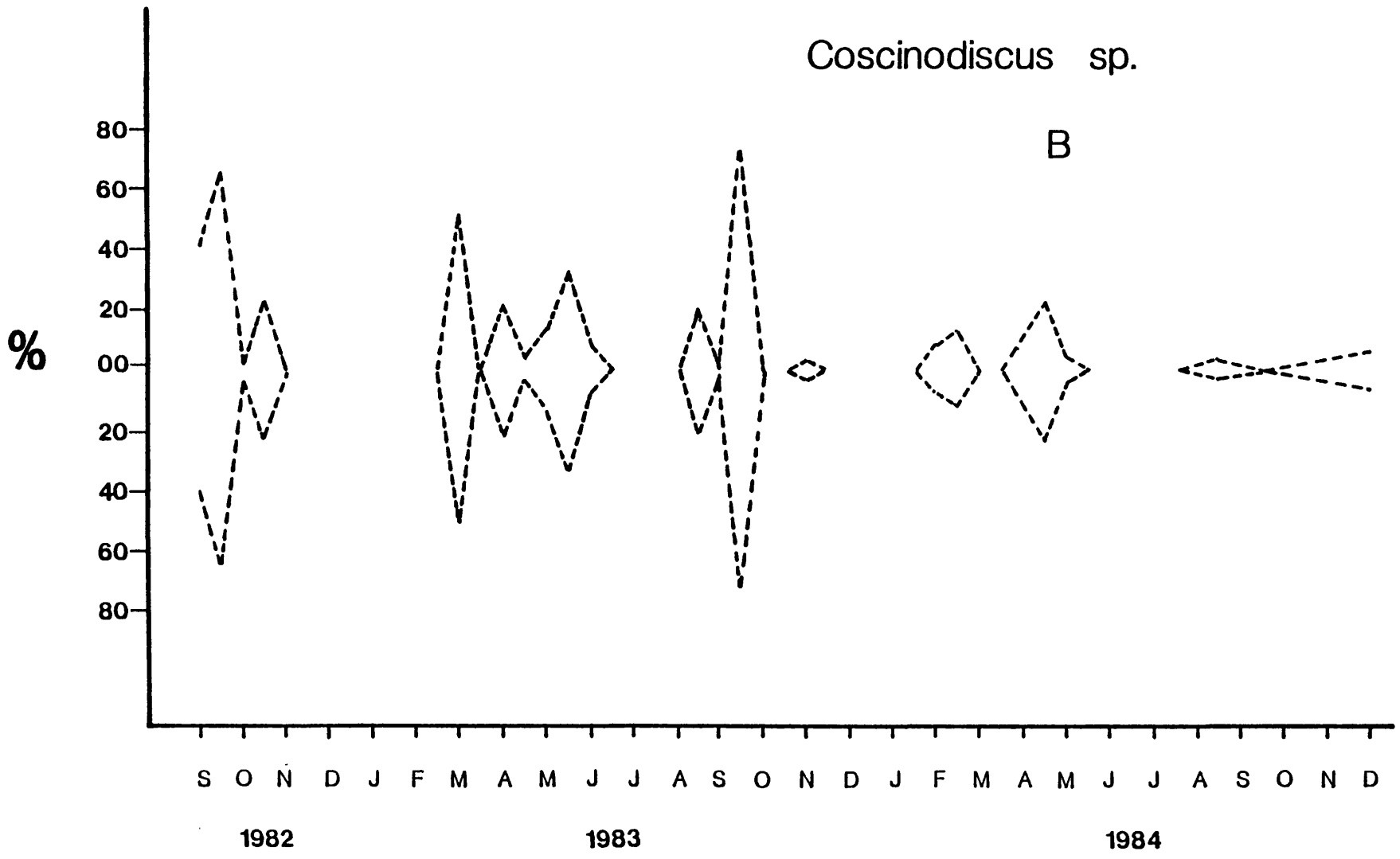

Coscinodiscus sp.
A
%
60
40
20
00
20
40
60
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

Figure 5.3.3 Dominance of Coscinodiscus sp.
(Station B)


Coscinodiscus sp.
B
%
80
60
40
20
00
20
40
60
80
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

that when the temporal variation in September, 1982 was very low (0.5%), Coscinodiscus sp. dominated the population, accounting for 65% of the total.

4. Chaetoceros sp.

During the sampling period, Chaetoceros sp. occurred mainly in the spring. It disappeared at other times of the year.

Station A (Figures 5.4.1 and 5.4.2)

From March, 1983 until August, 1983 the values of the temporal variations were between 0.5% to 1%, with a noticeable peak of 5% in May, 1983. The other period of Chaetoceros sp. growth took place from March, 1984 to August, 1984. The peaks of this period were of the same size, March, 1984 (1.5%); June, 1984 (0.5%) and August, 1984 (1%). See Figure 5.4.1.

At this station, the dominance of Chaetoceros sp. was not obvious, except in the sample of August, 1983 (44%). See Figure 5.4.2.

Station B (Figures 5.4.1 and 5.4.2)

From March, 1983 until August, 1983 the pattern of temporal variations was similar to that of Station A. It consisted mainly of low peaks of ca. 0.5%. The exception was the sample of May, 1983 with a value of 10%. This pattern was repeated from March to September, 1984. The highest peak in 1984 was in May, with a value of 12.5%. At this station Chaetoceros sp. dominated the phytoplankton population in May, 1983 and May, 1984 when it formed 30% and 63% of the population, respectively. When compared with the two previous figures, it can be seen that the values of temporal variations (Fig. 5.4.1, A) in May,

Figure 5.4.1 Temporal variation of Chaetoceros sp.
(Stations A and B)

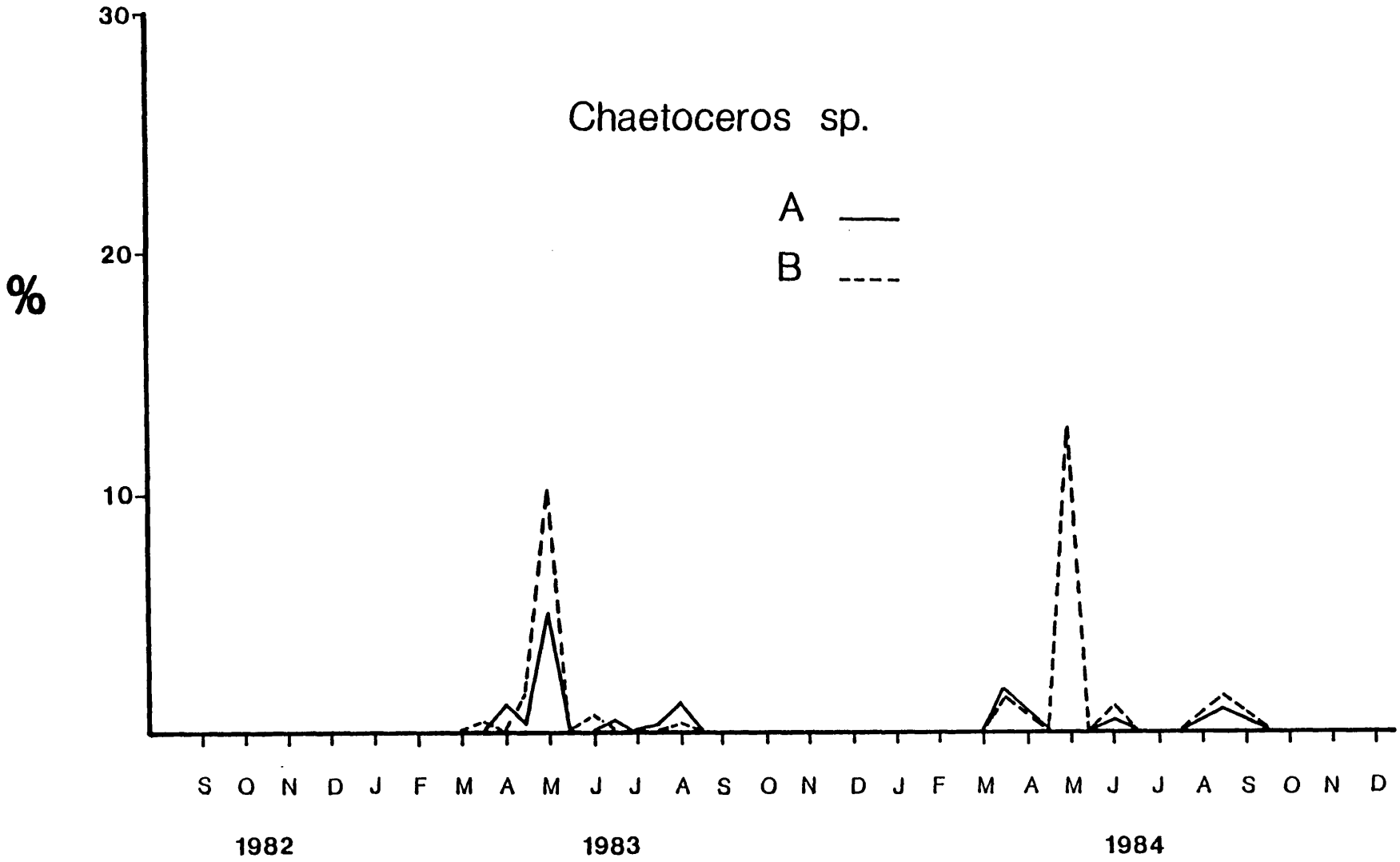
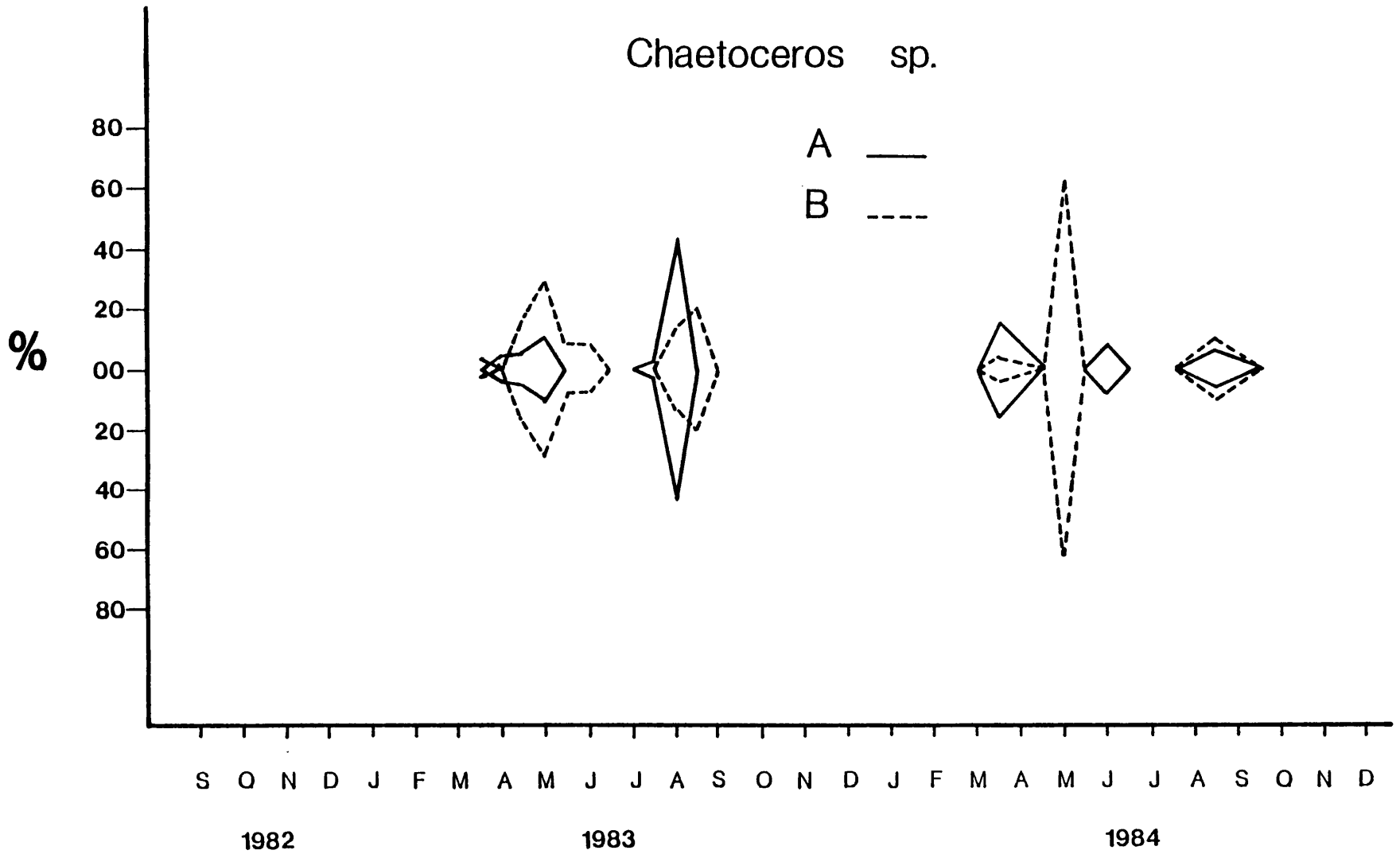

Chaetoceros sp.
A
B
%
30
20
10
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

Figure 5.4.2 Dominance of Chaetoceros sp.
(Stations A and B)


Chaetoceros sp.
A
B
%
80
60
40
20
00
20
40
60
80
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

1983 (5%) and August, 1983 (1%) did not correspond to the same extent in Figure 5.4.2, A. In this figure, the value in May, 1983 was 10%, while that of August, 1983 was 43%.

5. Rhizosolenia hebetata

This species occurred in the spring and summer of 1983 and 1984.

Station A (Figures 5.5.1 and 5.5.2)

The temporal variation (Fig. 5.5.1, A) featured four different peaks. This species first occurred in April, 1983 (18%); then in May, 1983 (4%); followed by April, 1984 (1%) and June, 1984 (1.5%). R. hebetata dominated the population in April, 1983 (47%) and May, 1983 (34%). It represented 27% of the population in June, 1984.

Station B (Figures 5.5.1 and 5.5.2)

In 1983, the temporal variation at this station was different from that of Station A (Fig. 5.5.1, A). It started in March, 1983 (1.5%) and maintained the same level in April and June, 1983. The highest peak was in July, 1983 which reached 10.5%. In 1984 it was generally lower than that of 1983 with a highest value of 1.5% in March. At this station, R. hebetata dominated the population on one occasion only. This was in July, 1983, reaching 30% (Fig. 5.5.2).

6. Melosira moniliformis

The major outburst of this species occurred in the early summer of 1983. It disappeared almost completely during the other seasons.

Figure 5.5.1 Temporal variation of Rhizosolenia hebetata
(Stations A and B)

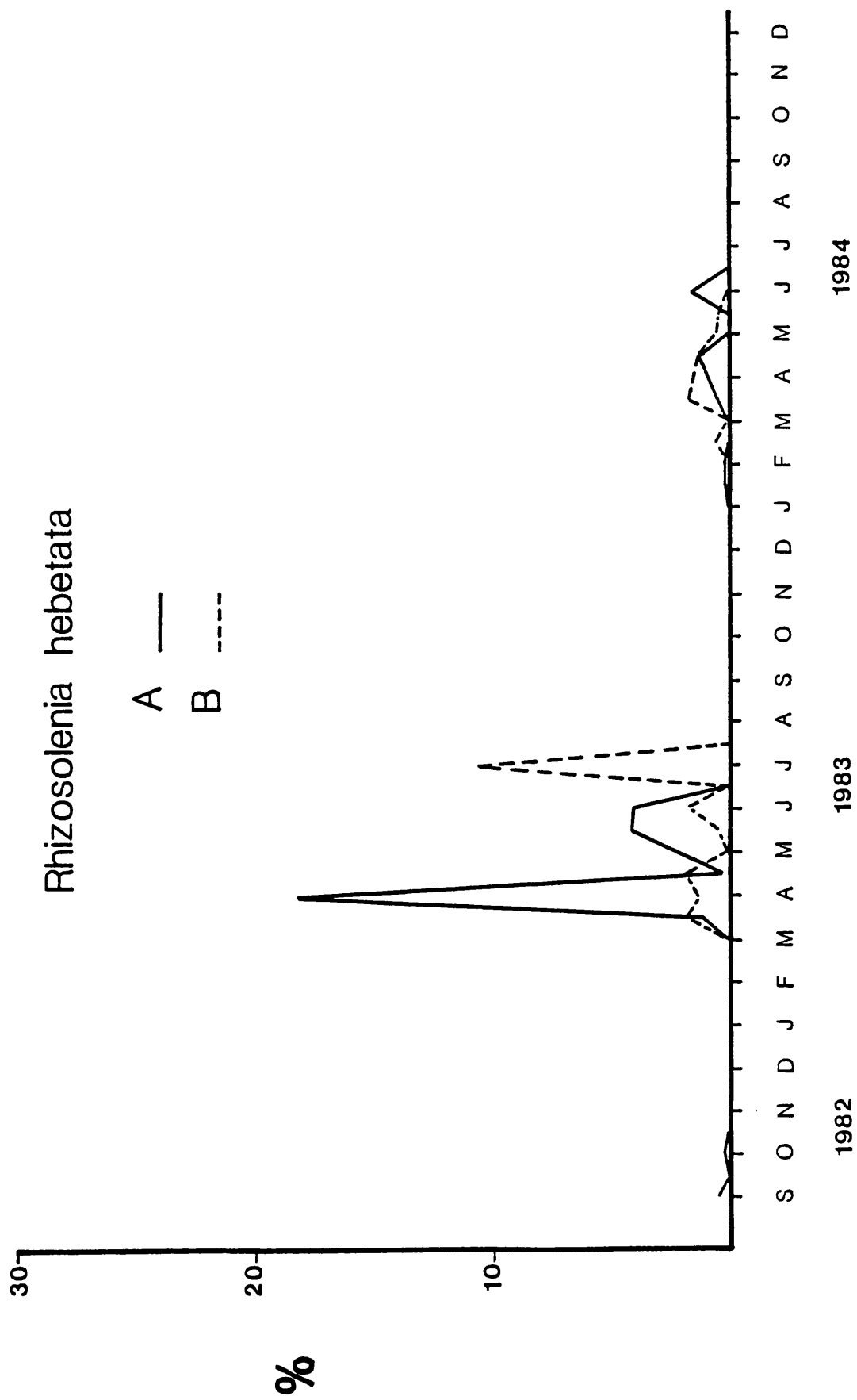
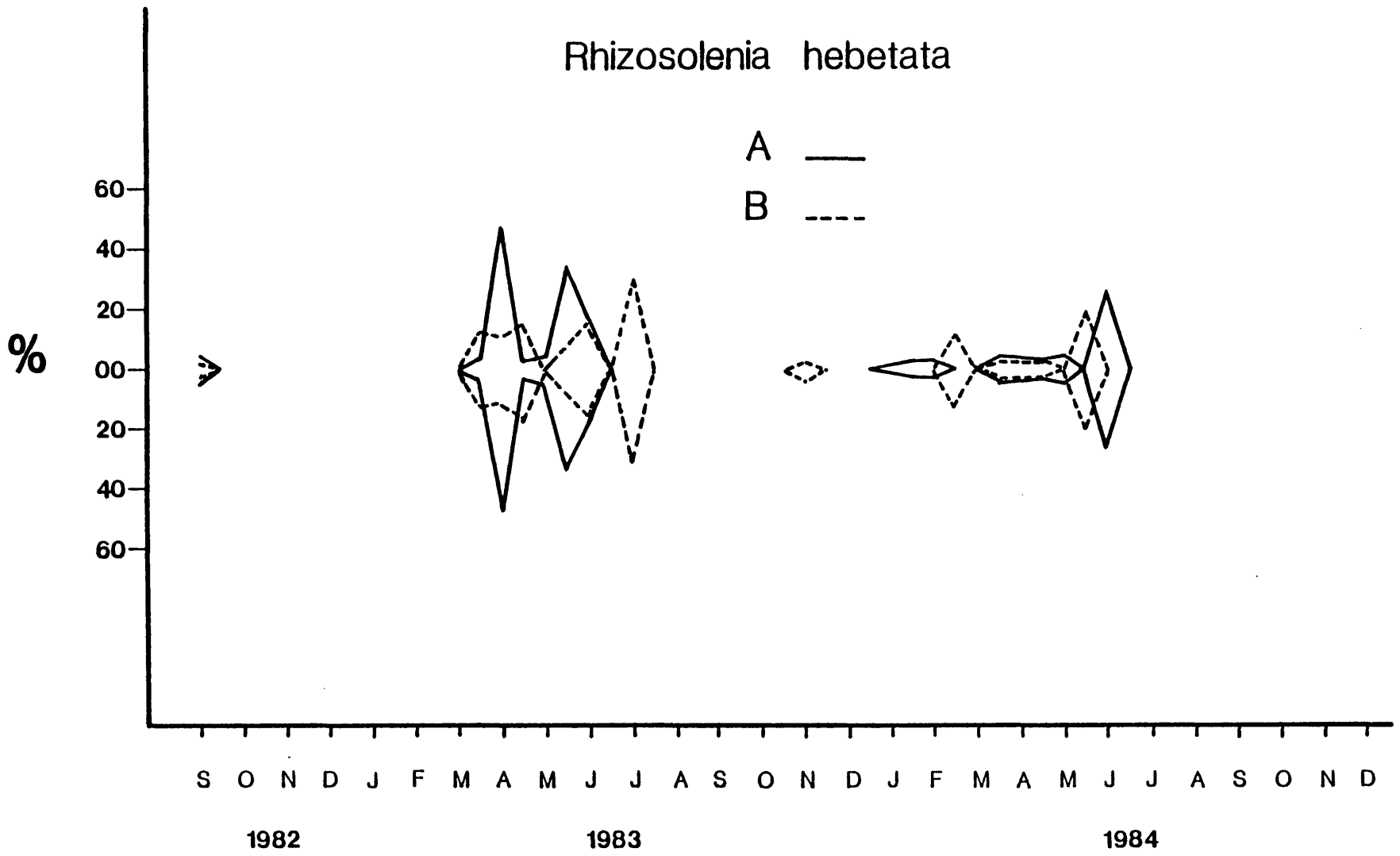

Rhizosolenia hebetata
A
B
%
30
20
10
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

Figure 5.5.2 Dominance of Rhizosolenia hebetata
(Stations A and B)


Rhizosolenia hebetata
A
B
%
60
40
20
00
20
40
60
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

Station A (Figures 5.6.1 and 5.6.2)

The temporal variation of this species was characterized by one major peak which took place in early June, 1983 reaching 45%. Melosira moniliformis accounted for 26% of the population of September, 1982 and 58% in June, 1983.

Station B (Figures 5.6.1 and 5.6.2)

At this station the temporal variation exhibited the same pattern as that of Station A. The major peak occurred at the same time in June, 1983 but reaching only 9% (Fig. 5.6.1). At this station, Melosira moniliformis dominated only once which was in June, 1983. On that occasion it accounted for 79% of the population.

7. Prorocentrum micans

P. micans was the most important dinoflagellate which occurred in Swansea Bay during the present study and dominated the phytoplankton population at one stage.

Station A (Figures 5.7.1 and 5.7.2)

At this station, P. micans occurred from April until October, 1983 and from April until December, 1984. The values of temporal variation were 1.5% in April, 1983; 4% in June, 1983; 1.5% in July, 1983; 2% in September, 1983; 1% in May, 1984; 3% in July, 1984; 6% in August, 1984; with the highest value in September, 1984 (9%). See Figure 5.7.1.

P. micans was dominant in August, 1983 (30%); May, 1984 (30%) and July, 1984 (40%). See Figure 5.7.2.

By comparing Figures 5.7.1 and 5.7.2 it can be noticed that when the temporal variation values were 3% (July, 1984); 6% (August, 1984)

Figure 5.6.1 Temporal variation of Melosira moniliformis
(Stations A and B)

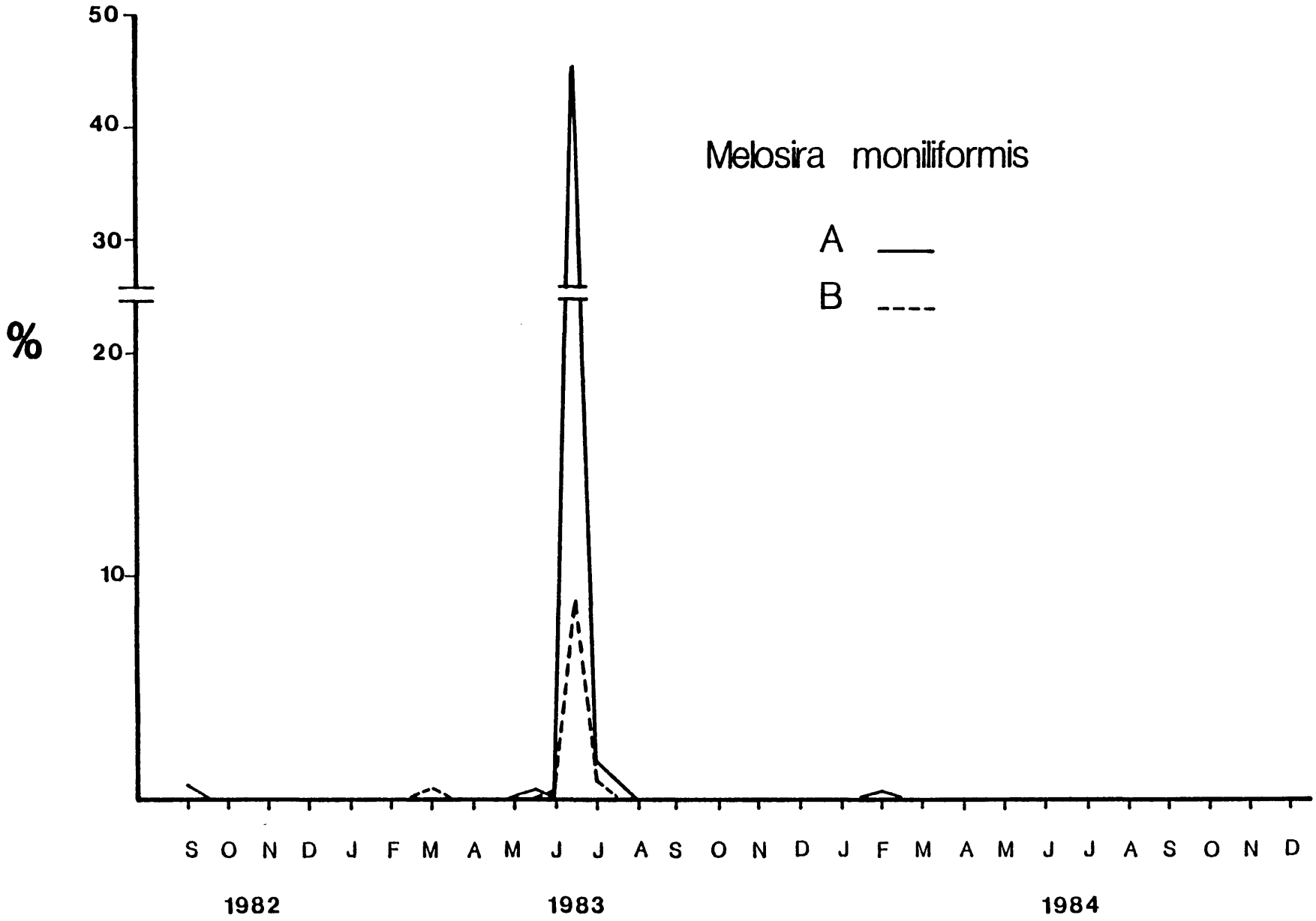

50
40
30
20
10
%
Melosira moniliformis
A
B
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

Figure 5.6.2 Dominance of Melosira moniliformis
(Stations A and B)

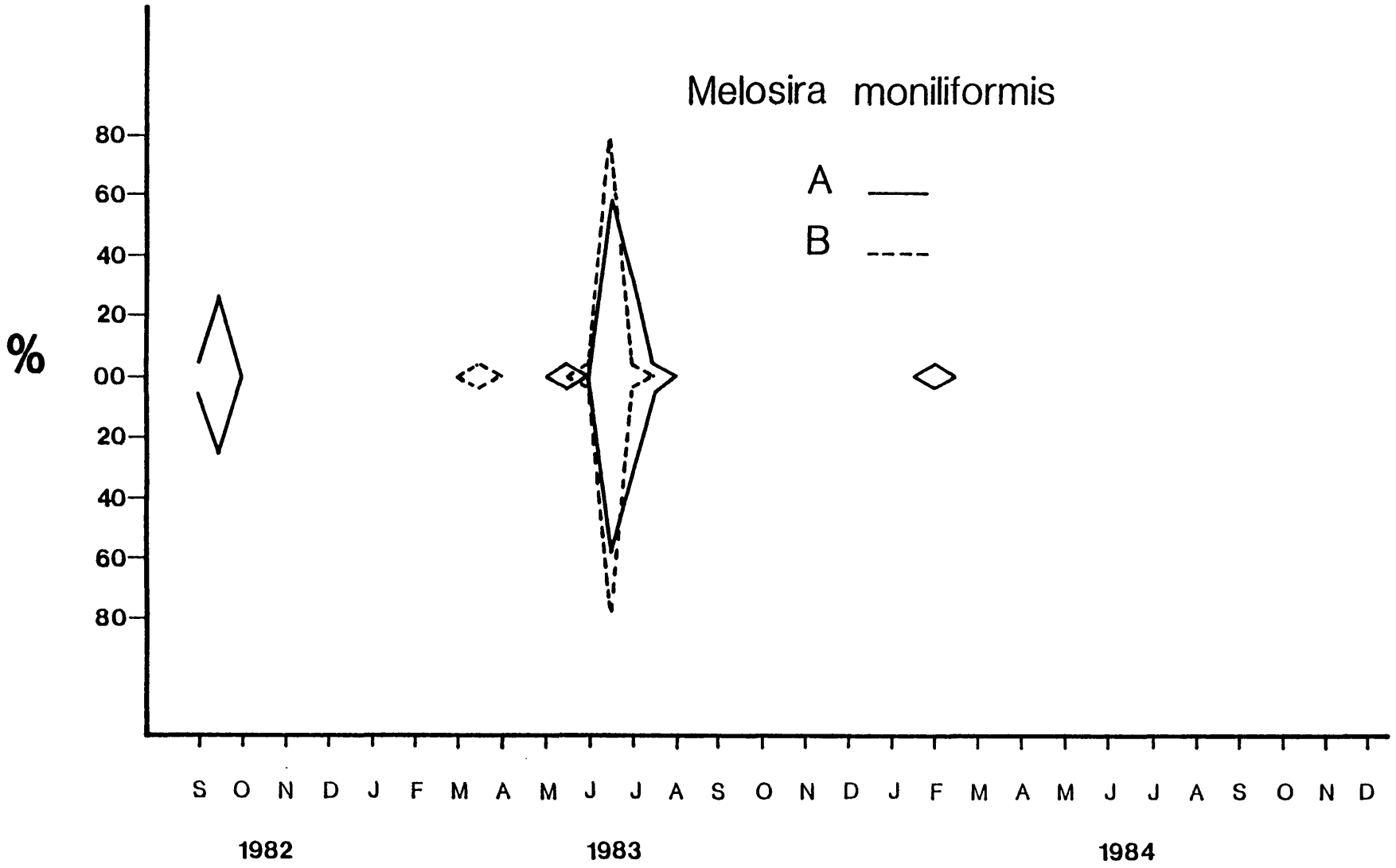

Melosira moniliformis
A
B
%
80
60
40
20
00
20
40
60
80
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

Figure 5.7.1 Temporal variation of Prorocentrum micans
(Stations A and B)

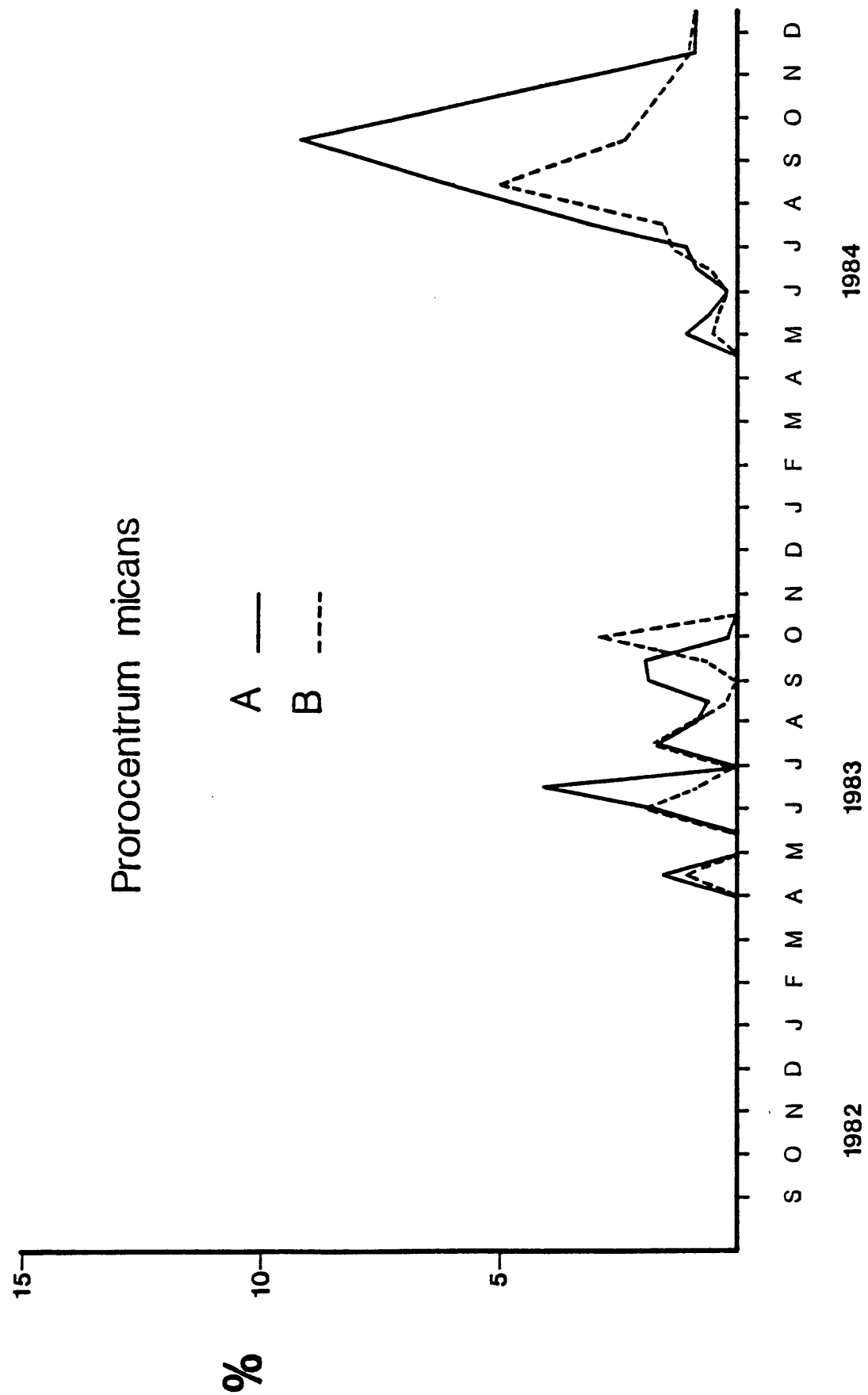
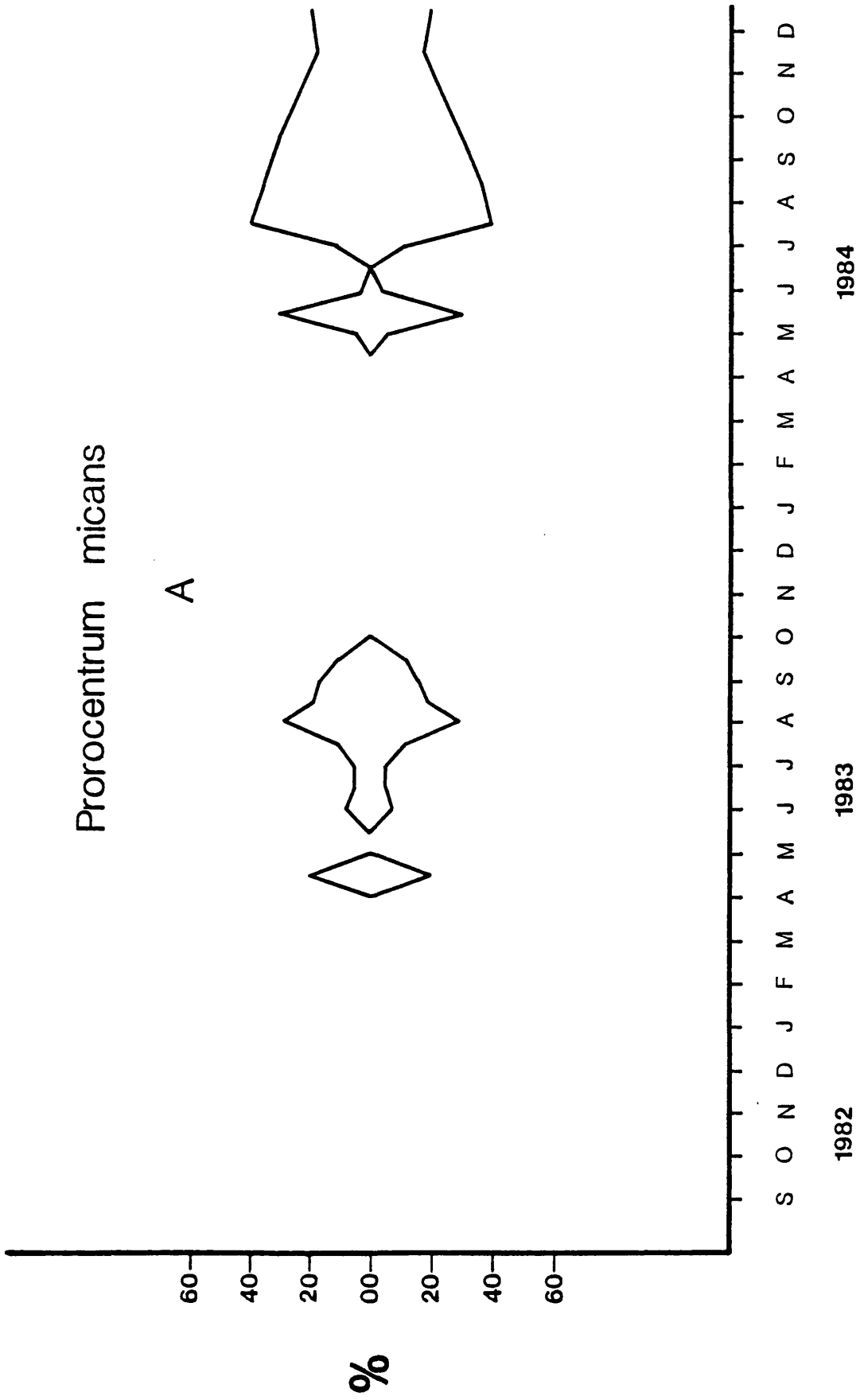

Prorocentrum micans
A ——
B ----
%
15
10
5
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

Figure 5.7.2 Dominance of *Prorocentrum micans*
(Station A)


Prorocentrum micans
A
%
60
40
20
00
20
40
60
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

and 9% (September, 1984) the dominance values were 40%, 36% and 30% respectively.

Station B (Figures 5.7.1 and 5.7.3)

At this station, P. micans occurred from April until October, 1983 and from May until December, 1984. The values of temporal variation were 1% in April, 1983; 2% in June, 1983; 2% in July, 1983; 3% in October, 1983, 1% in May, 1984 and the highest value was 5% in August, 1984.

P. micans contributed 29% of August, 1983 population. It dominated the population of July, 1984 (40%) and August, 1984 (33%). There was contrariety between Figure 5.7.1 and 5.7.3. It can be noticed that when the temporal variation values were 1.5% (July, 1984); 5% (August, 1984) and 2.5% (September, 1984) the dominating values were 40%, 33% and 30% respectively.

8. Rhizosolenia shrubsolei

Rhizosolenia shrubsolei is one member of the phytoplankton which occurred in the summer months only.

Station A (Figures 5.8.1 and 5.8.2)

When the temporal variation of R. shrubsolei is considered, it can be mentioned that the individuals of this species occurred more in the summer of 1983 than in the summer of 1984 (Fig. 5.8.1). In July, 1983 it reached 9.5%, while in June, 1984 it was only 1%.

This species dominated the phytoplankton population on one occasion only, which was in July 1983 when it reached 60%.

Figure 5.7.3 Dominance of Prorocentrum micans
(Station B)

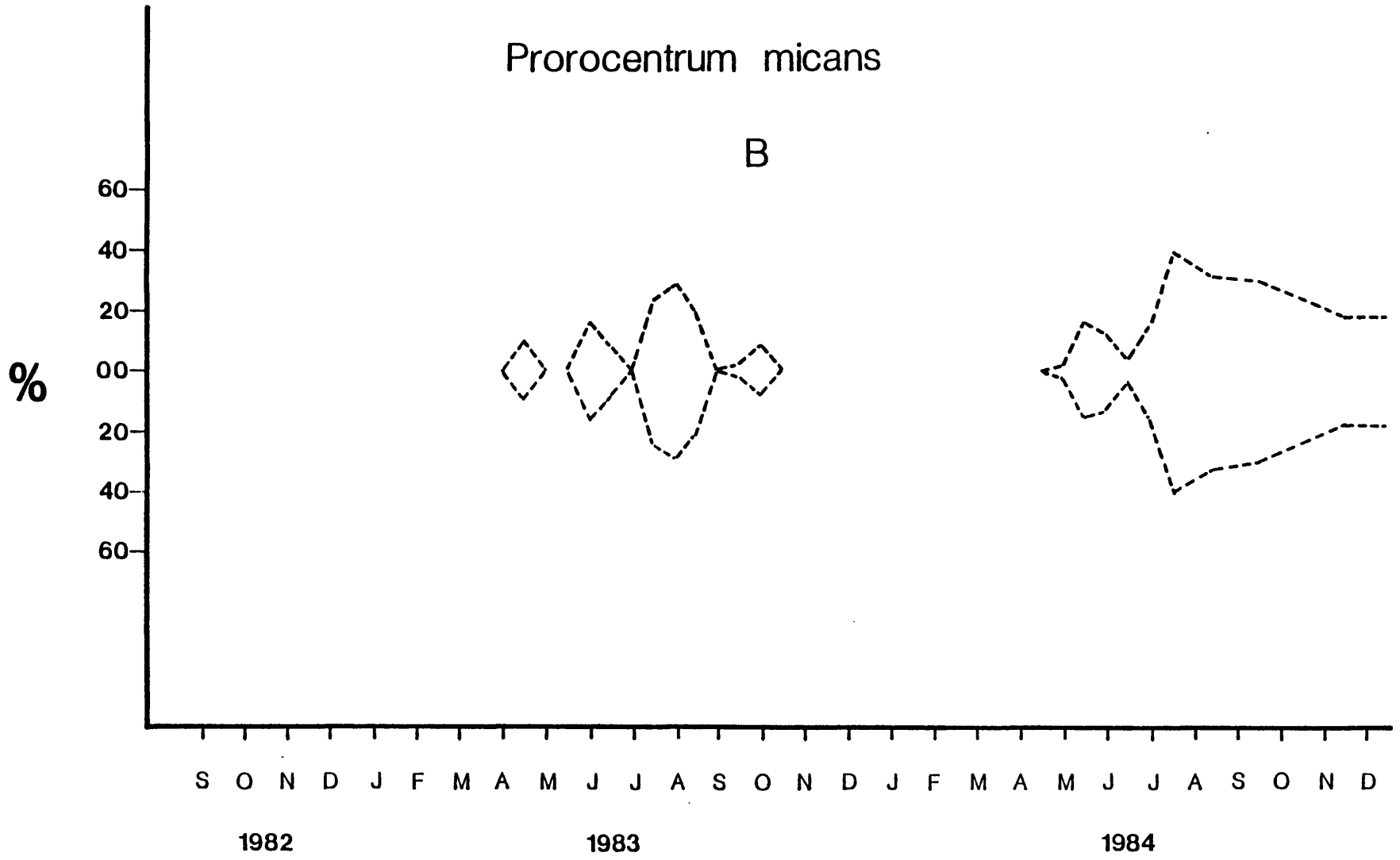

Prorocentrum micans
B
%
60
40
20
00
20
40
60
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

Figure 5.8.1 Temporal variation of Rhizosolenia shrubsolei
(Stations A and B)

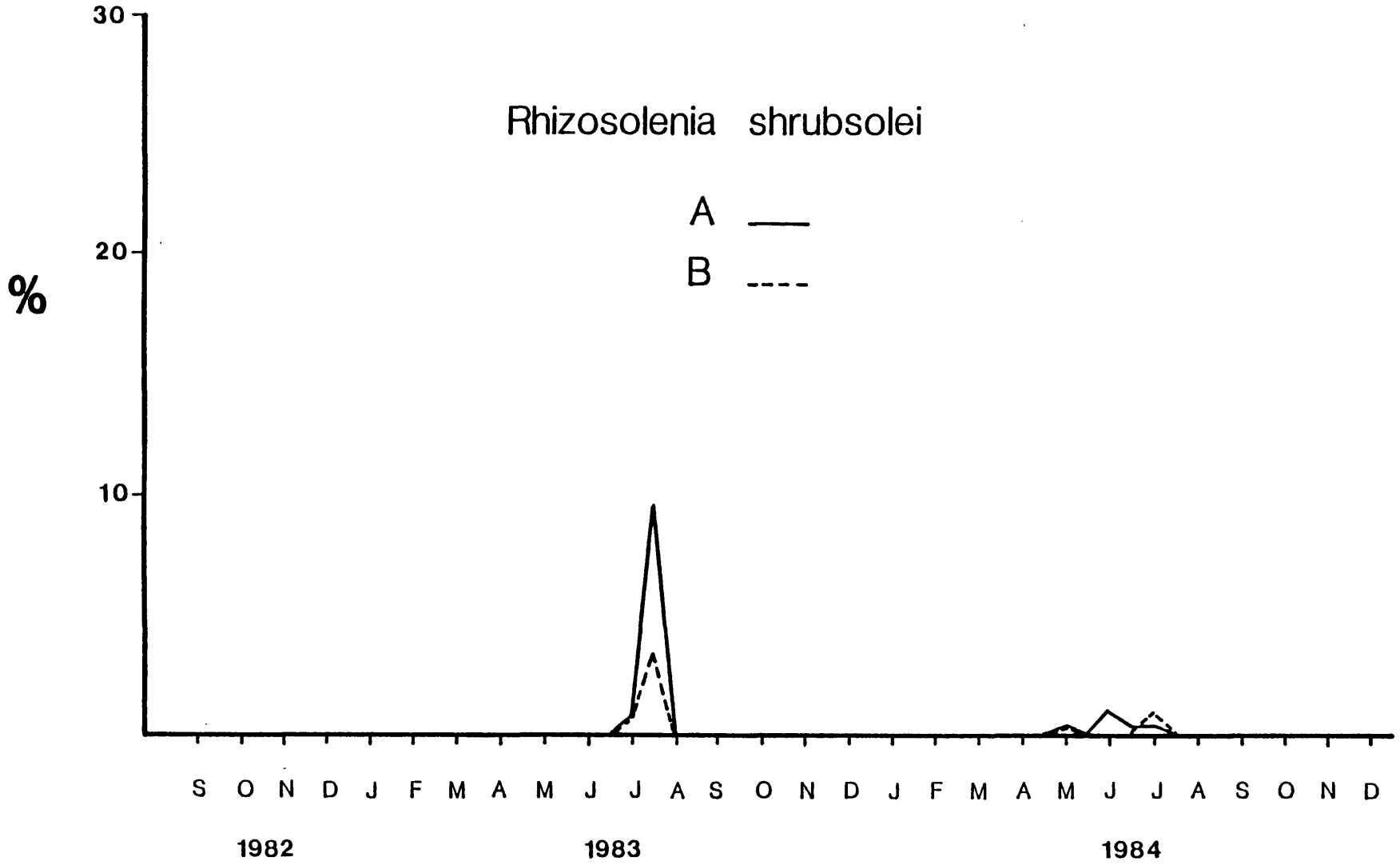
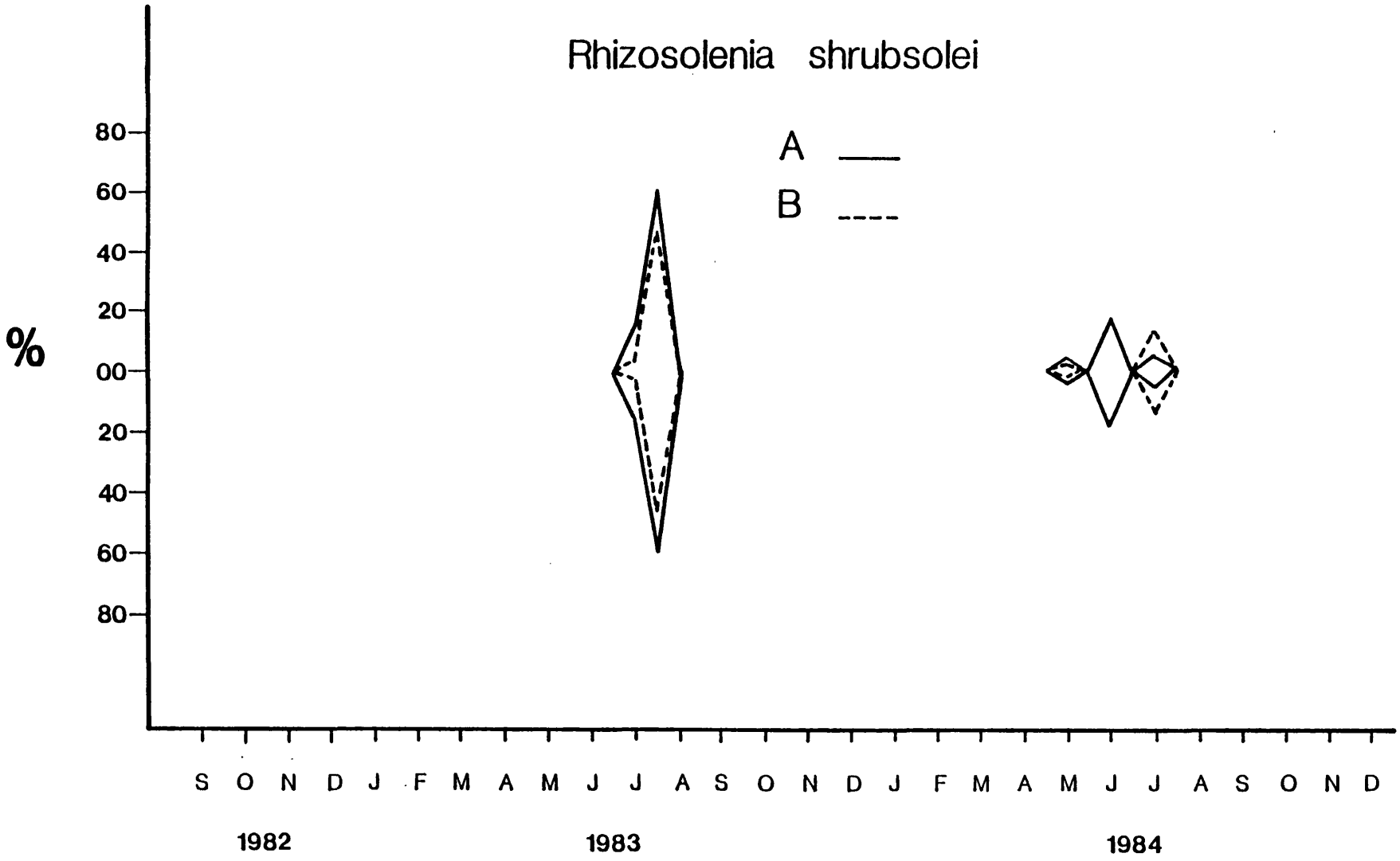

Rhizosolenia shrubsolei
A
B
30
20
10
%
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

Figure 5.8.2 Dominance of Rhizosolenia shrubsolei
(Stations A and B)


Rhizosolenia shrubsolei
A
B
%
80
60
40
20
00
20
40
60
80
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

Station B (Figures 5.8.1 and 5.8.2)

In this station, R. shrubsolei took the same pattern as was seen in Station A, but at a lower level. In fact, the highest peak in this station was 3.5% in July, 1983. It occurred at the same time in 1984, but it only reached 1%. R. shrubsolei dominated the phytoplankton in July, 1983 when it accounted for 46% of the population (Fig. 5.8.2).

9. Skeletonema costatum

Station A (Figures 5.9.1 and 5.9.2)

S. costatum occurred in low numbers in 1983 (Fig. 5.9.1). This occurrence extended from April to July, 1983. In May and June, 1983 it was 1%. This pattern was changed in 1984 when the value reached 15.5% in April. S. costatum was the dominant species in April, 1984. In the sample of that month, it represented 38% of the population (Fig. 5.9.2)

Station B (Figures 5.9.1 and 5.9.2)

The pattern of temporal variation which took place in Station A was similar at this station. The highest value of 1983 was 1% in May. In the growth season of 1984, the highest value was 51% in April (Fig. 5.9.1). The contribution of S. costatum to the population of April, 1984 was 29%

10. Thalassionema nitzschioides

Station A (Figures 5.10.1 and 5.10.2)

This species was recorded on few occasions during this study. It occurred in low densities in April, 1983 (1%) and May, 1983 (2%). It was also found in the spring of 1984 in low densities of 1% (February) and 0.5% in March. Although it occurred in negligible numbers in

Figure 5.9.1 Temporal variation of Skeletonema costatum
(Stations A and B)

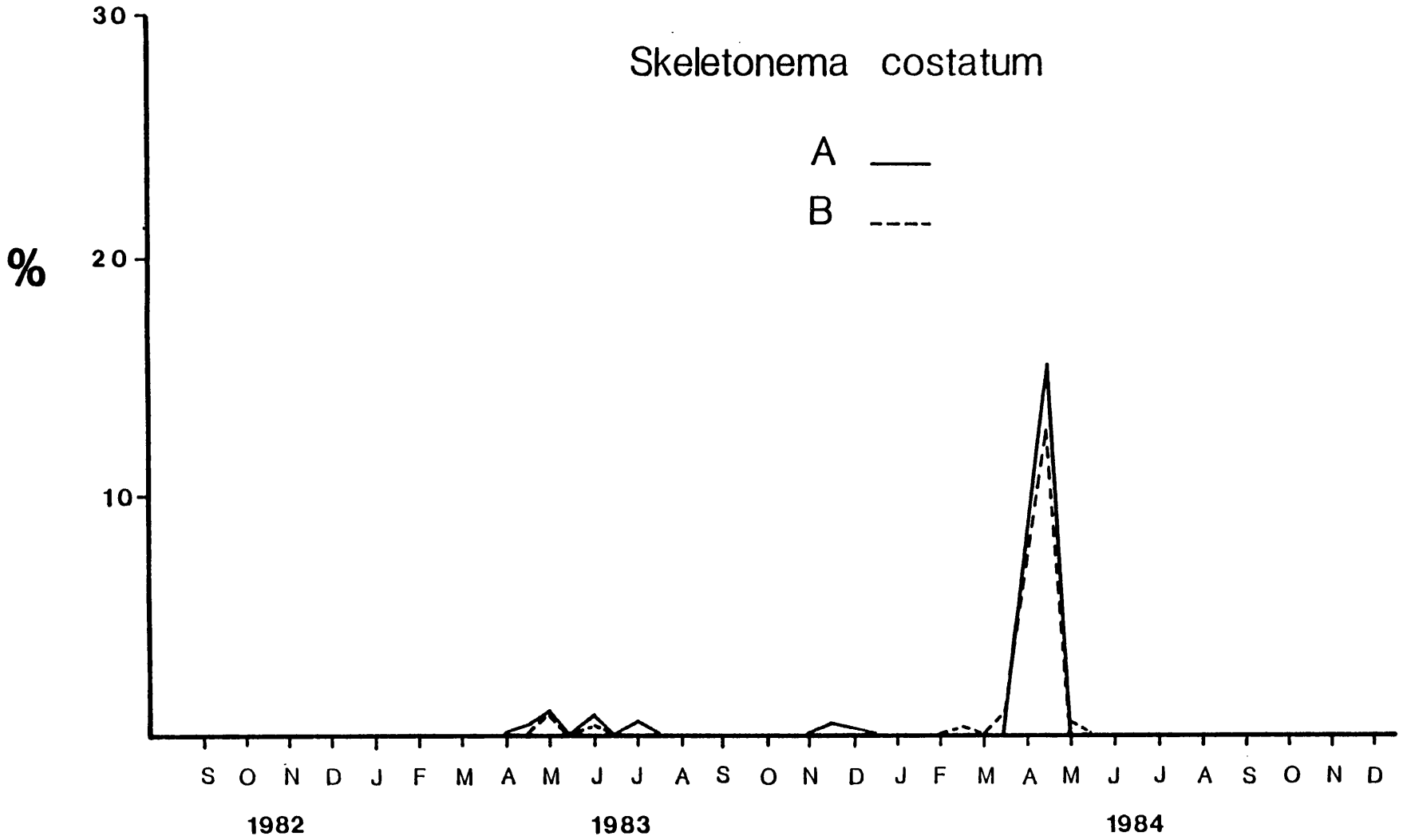

Skeletonema costatum
A
B
%
30
20
10
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

Figure 5.9.2 Dominance of Skeletonema costatum
(Stations A and B)

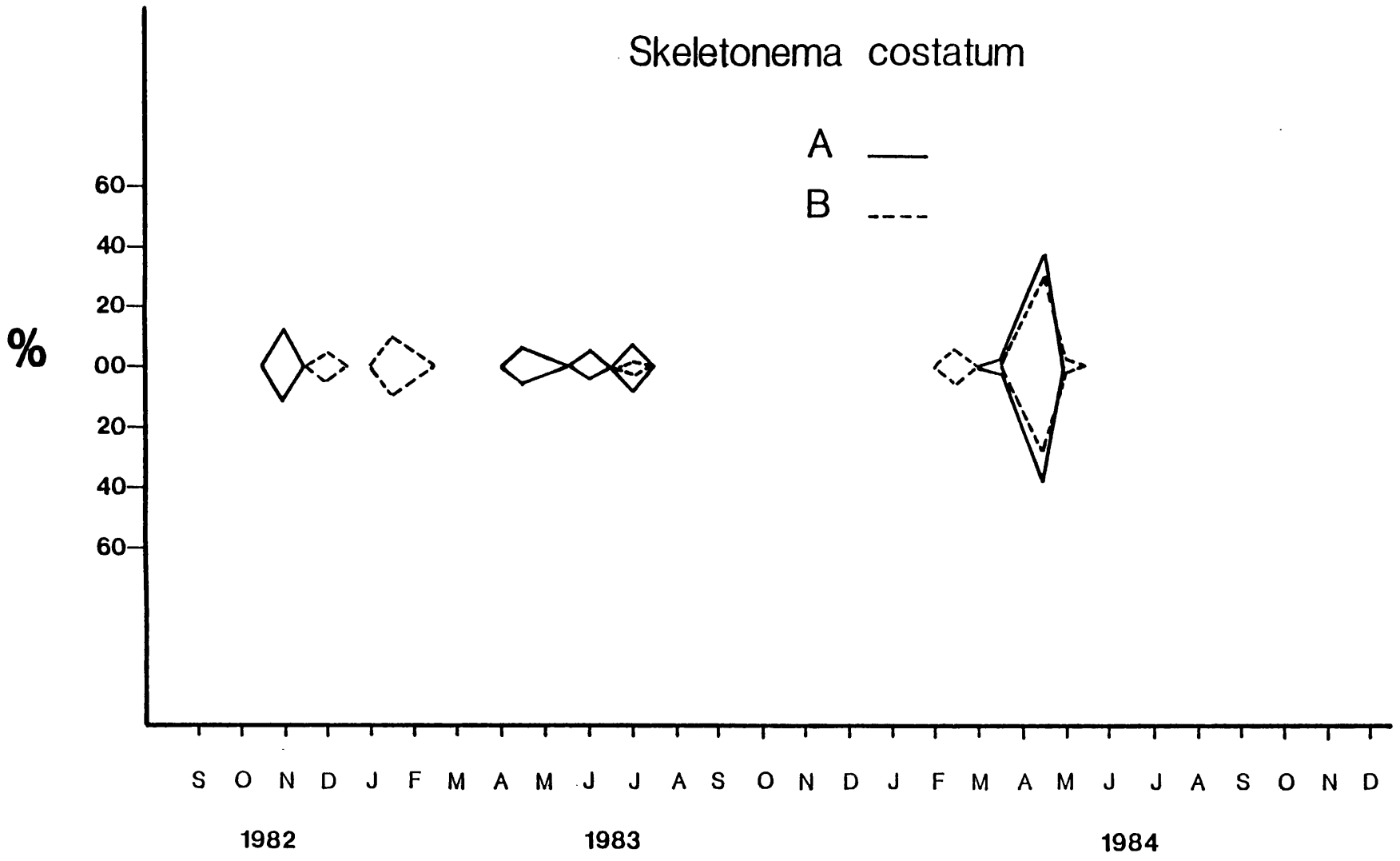

Skeletonema costatum
A
B
%
60
40
20
00
20
40
60
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

Figure 5.10.1 Temporal variation of Thalassionema nitzschioides (Stations A and B)

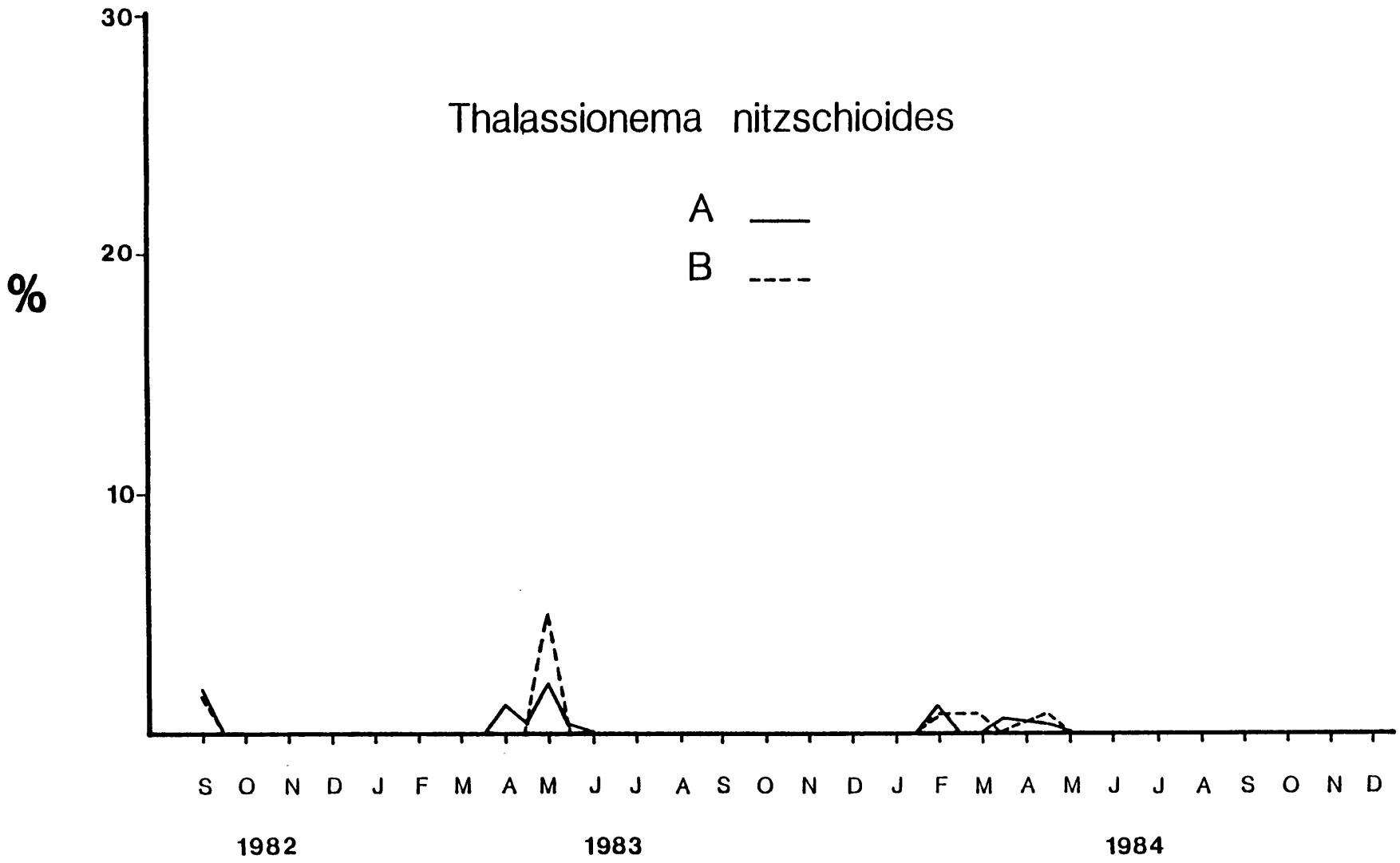
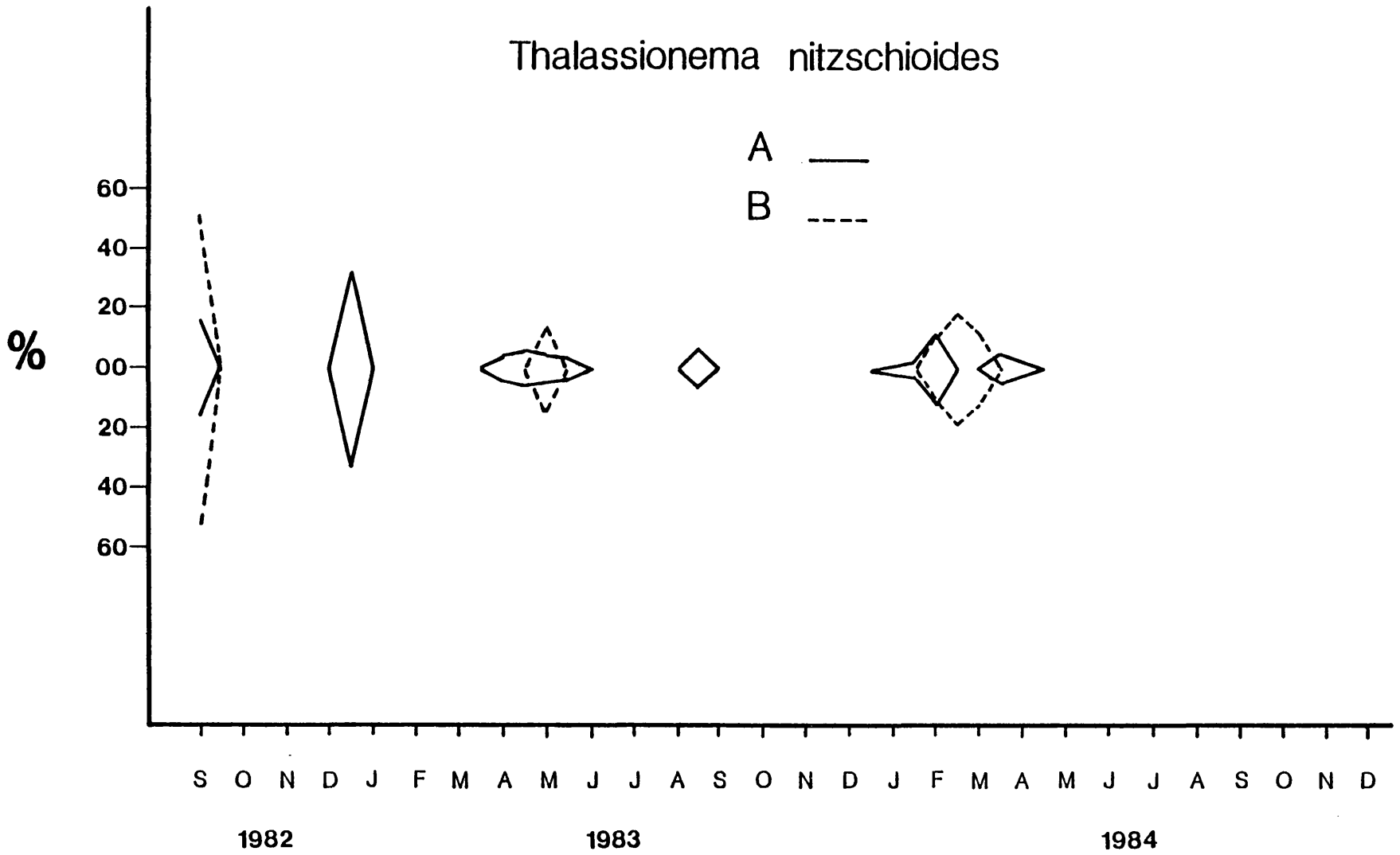

Thalassionema nitzschioides
A
B
%
30
20
10
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

Figure 5.10.2 Dominance of Thalassionema nitzschioides (Stations A and B)


Thalassionema nitzschioides
A
B
%
60
40
20
00
20
40
60
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

December, 1982, it dominated the population of that month with a value of 33%.

Station B (Figures 5.10.1 and 5.10.2)

The temporal variation in this station was characterized by low values throughout the occurrence periods (Fig. 5.10.1). An exception to this pattern was the relatively high peak of 5% in May, 1983. The second highest value was in September, 1982 (1.5%). In that month, T. nitzschioides was dominant, representing 52% of the population (Fig. 5.10.2).

11. Rhizosolenia delicatula

Station A (Figures 5.11.1 and 5.11.2)

Throughout the sampling period, R. delicatula occurred only once, in June, 1984. It accounted for 92% of the highest recorded population (Fig. 5.11.1). On that occasion it represented 95% of the population (Fig. 5.11.2).

Station B (Figures 5.11.1 and 5.11.2)

At this station, the temporal variation took the same pattern as that of Station A. There was only one peak which reached 10% in June, 1984. When it occurred in June, 1984 R. delicatula dominated the population and accounted for 70%.

12. Thalassiosira gravida

Station A (Figures 5.12.1 and 5.12.2)

The temporal variation of T. gravida at this station was restricted to April and May, 1984. The highest value was in May (4.5%). On the

Figure 5.11.1 Temporal variation of Rhizosolenia delicatula
(Stations A and B)

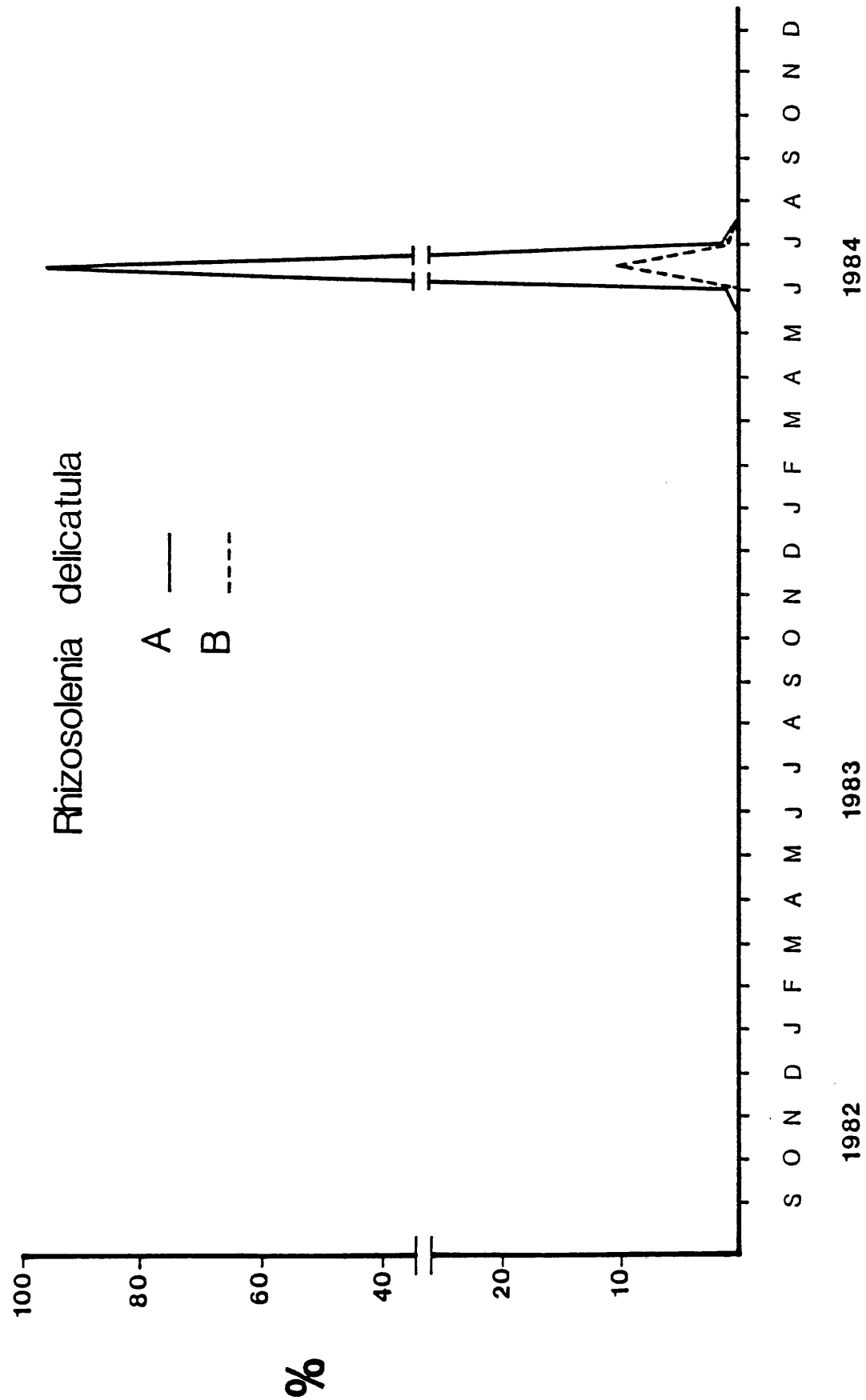

Rhizosolenia delicatula
A
B
%
100
80
60
40
20
10
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

Figure 5.11.2 Dominance of Rhizosolenia delicatula
(Stations A and B)

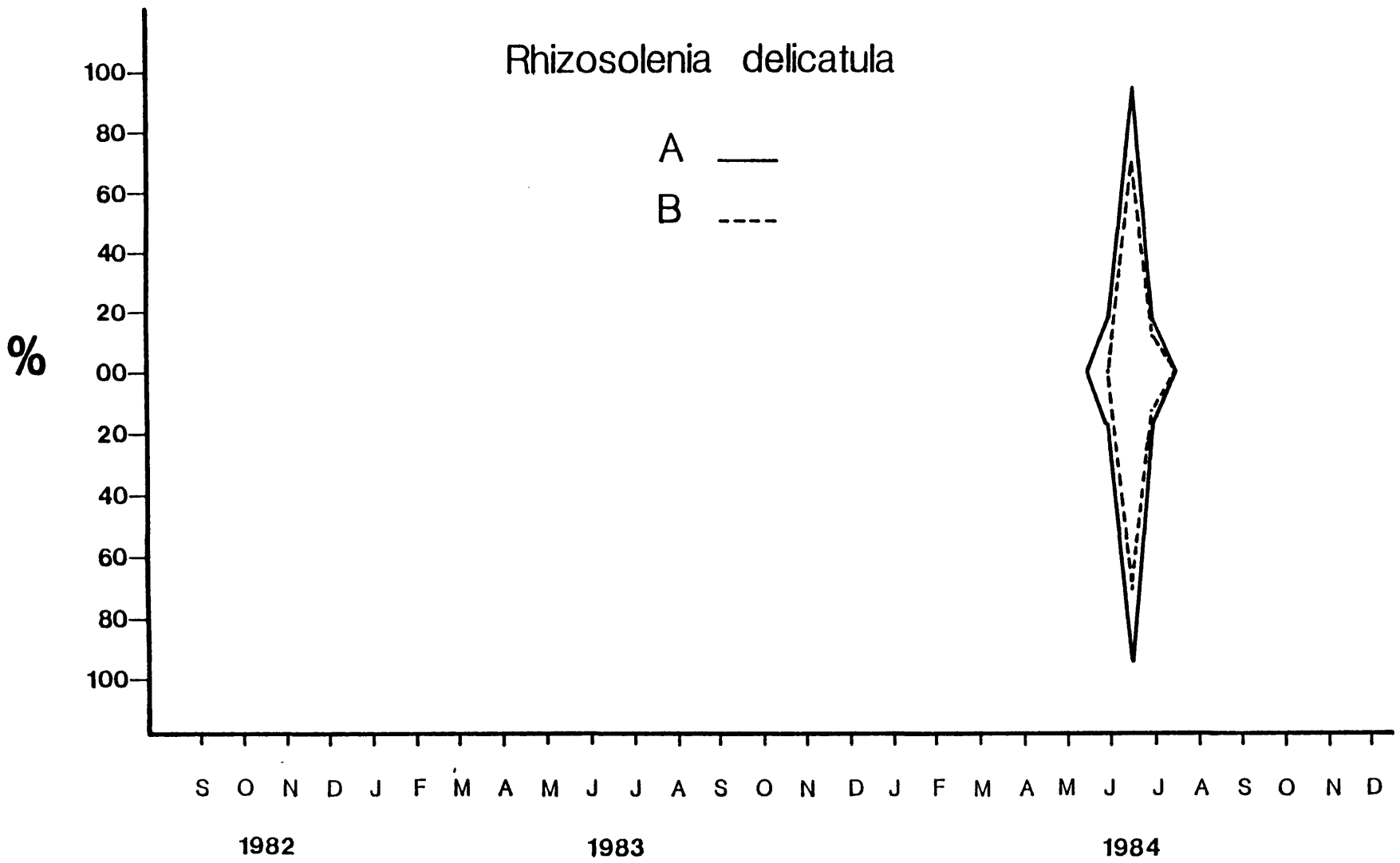

Rhizosolenia delicatula
A ——
B ----
%
100
80
60
40
20
00
20
40
60
80
100
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

Figure 5.12.1 Temporal variation of Thalassiosira gravida
(Stations A and B)

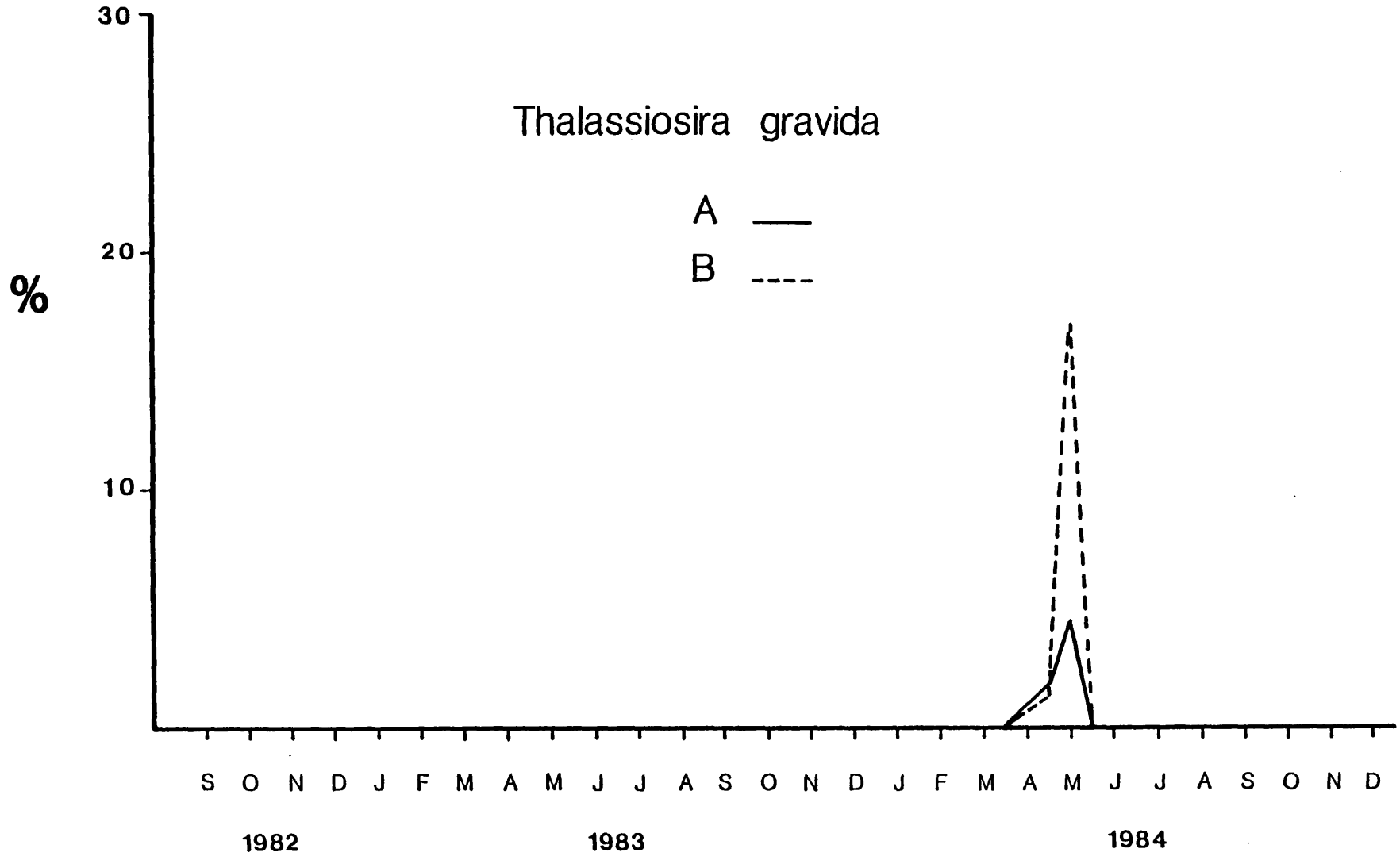
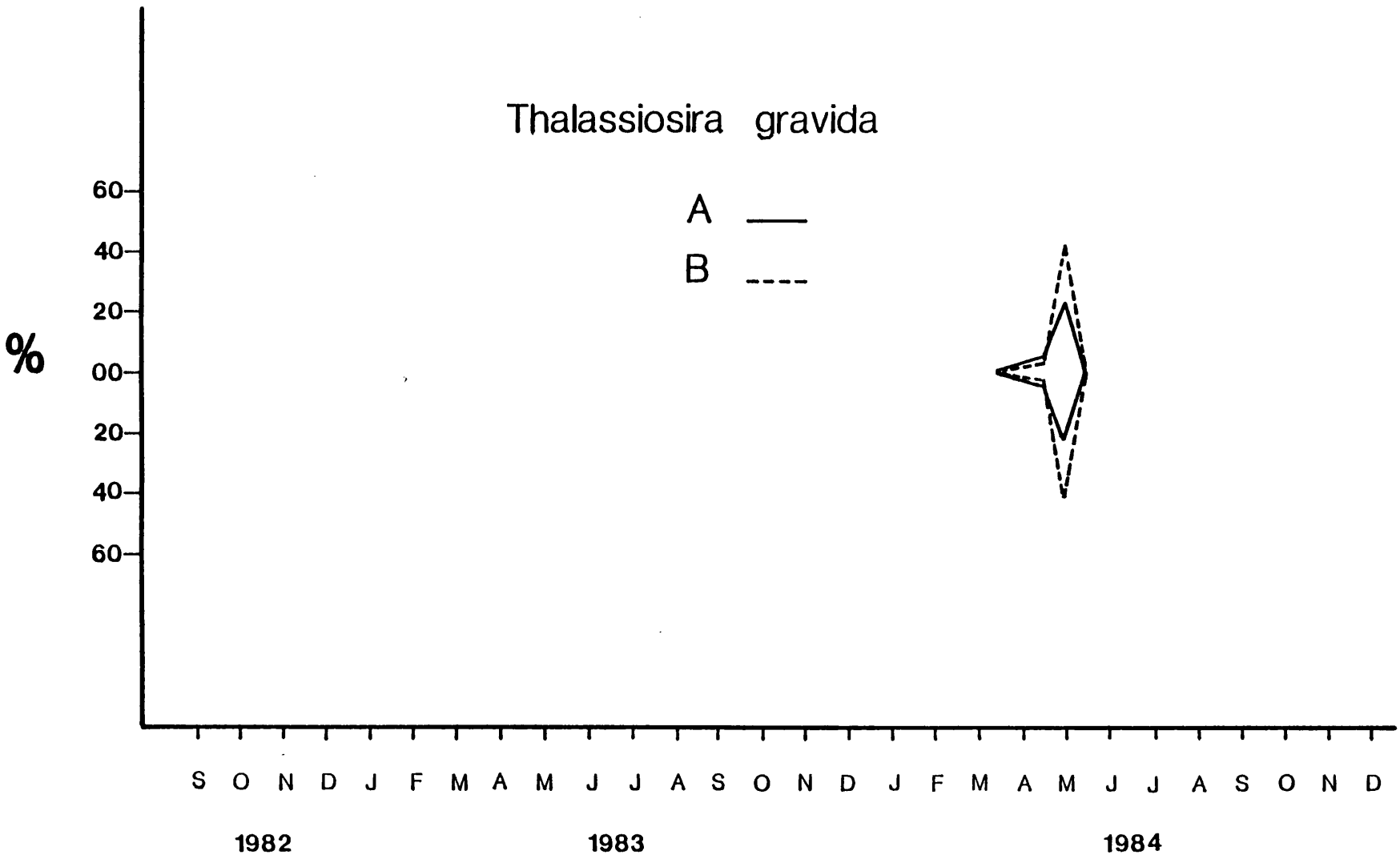

Thalassiosira gravida
A
B
%
30
20
10
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

Figure 5.12.2 Dominance of Thalassiosira gravida
(Stations A and B)


Thalassiosira gravida
A
B
%
60
40
20
00
20
40
60
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

other hand, this species accounted for 22% of the May population.

Station B (Figures 5.12.1 and 5.12.2)

In contrast to the highest value of temporal variation in Station A, the highest value at this station was 17%. This value occurred in May, 1984 (Fig. 5.12.1). The individuals of this species were dominant in May, 1984. They represented 42% of that month's population (Fig. 5.12.2).

13. Phaeocystis sp.

It is almost certain that Phaeocystis was responsible for at least part of the production in Swansea Bay, especially in the summer. This species was recorded at peak levels in the samples of May-June, 1984. Due to its delicate and slimy structure, it broke down and clogged the towing net which made it very difficult to quantify its presence in the net sample.

DISCUSSION

There is a continuous change in the biotic and abiotic environmental factors througout the year. This change may take the shape of a seasonal cycle or just an occasional pulse which may occur at any time due to one reason or another. The change in the environment which is caused by a single factor or a combination of several factors may affect the population of certain species or the community as a whole.

The cyclic and seasonal variations in the environmental conditions of a climatic region produce a characteristic seasonal pattern in the

phytoplankton of such a region. The seasonal growth pattern which is characteristic of the temperate regions has been recorded from coastal waters in Britain by Atkins (1923), Harvey (1933), Barry (1963), Pearce (1967), Dooley (1973), Vogelmann (1980) and Sexton (1985).

The phytoplankton which occurred and dominated the population in Swansea Bay was divided into different groups according to the frequency of occurrence:

A. A group of continuous occurrence;
B. A group of discontinuous occurrence;
C. Species with one major occurrence.

A. A group of continuous occurrence

This group contains the species which occurred throughout the year, including Bacillaria paxillifer, Biddulphia sinensis and Coscinodiscus sp..

1. Bacillaria paxillifer (Figures 5.1.1 to 5.1.4)

Bacillaria paxillifer was found to be neritic (Lebour, 1929). This species has been recorded around the Gower peninsula coasts by Paulraj (1974), Vogelmann (1980) and Sexton (1985). It was found to be one of the dominant species which occurred throughout the year.

In the present study, a comparison of the environmental conditions which preceded the major peaks at Station A (31.3.1983 and 19.10.1983) and B (11.7.1983 and 20.3.1984) was made (Table 5.1)

When the conditions of A (4.3.1983) were compared with those of B (6.3.1984), it was found that the recorded environmental conditions at both stations were similar. These conditions are usually

Table 5.1 Environmental factors measured during the growth of *Bacillaria paxillifer*

FACTOR	STATION A		STATION B	
	4.3.83	10.10.83	23.6.83	6.3.84
Salinity (‰)	28.2	28.6	29	29.4
Temperature (°C)	5.5	14.3	15.5	7
Nitrate (µg at Nl^{-1})	33.57	19.3	6.7	47.1
Nitrite (µg at Nl^{-1})	0.2	2.6	0.24	0.075
Ammonia (µg at Nl^{-1})	0.21	0.07	0	0.03
Silicate (µg at Sil^{-1})	16.4	11.8	0.47	16.95
Phosphate (µg at Pl^{-1})	0.83	0.99	0.1	1.03

characteristic of the spring season, i.e. a relatively low temperature accompanied by high concentrations of nutrients. In this case, it is possible that the combination of high nutrient concentrations and cool temperatures of the spring (5.5-8°C) may have triggered the growth of *B. paxillifer*. On the other hand, another group of *B. paxillifer* growth peaks was found in the sample of 11.7.1983 (B) and 19.10.1983 (A). The magnitude of the environmental conditions in this case was different from that of the spring growth (Table 5.1).

Firstly, the temperatures of the period which preceded the peaks were much higher than those of the spring. Secondly, the peak of *B. paxillifer* which occurred at Station A (19.10.1983) was preceded by relatively high levels of nutrient concentrations especially nitrite which reached very high concentraions (*ca.* 2.6 µg at Nl^{-1}).

The peak which took place in the summer (11.7.1983) in Station B

was preceded by very low nutrient concentrations especially of silicate. Temperature was the only factor which was as high as that of 19.10.1983 growth. It appears that temperature was the main factor which stimulated the growth of B. paxillifer in the late summer and autumn.

From all the points mentioned in the previous discussion and the possibilities which were suggested to explain the growth of B. paxillifer during the study period, it can be noticed that those possibilities were applied to explain the growth of B. paxillifer in hugely different conditions. The conditions varied from time to time during the periods which preceded the growth. Temperatures varied from 5.5 to 15.5°C, nitrate from 6.7 to 47.1 µg at Nl^{-1}, nitrite from 0.075 to 2.6 µg at Nl^{-1}, ammonia from 0 to 0.21 µg at Nl^{-1}, silicate from 0.47 to 16.95 µg at Sil^{-1} and phosphate 0.1 to 1.03 µg at Pl^{-1}.

To explain this continuous occurrence of B. paxillifer throughout the year two possibilities emerge: firstly, an unmeasured environmental factor or combination of factors may have been limiting the growth or, secondly, different physiological races of B. paxillifer may occur at different times of the year regulated mainly by the temperature (Vogelmann, 1980; Sexton, 1985). A spring race which is favoured by temperature around 5°C, and an autumn race with a temperature preference over the range 14-18°C.

2. Biddulphia sinensis (Figures 5.2.1 to 5.2.4)

Biddulphia sinensis is a major representative of the phytoplankton found in the waters around the British coasts. It has been recorded in the water samples of the Gower peninsula coasts by Paulraj (1974), Vogelmann (1980) and Sexton (1985).

Like Bacillaria paxillifer, Biddulphia sinensis was found to occur throughout the year.

In the present study, the seasonal variation pattern of Biddulphia sinensis was very similar to that of B. paxillifer (Figs. 5.1.1, 5.1.3, 5.2.1, 5.2.3). In Station A, the major peaks of growth occurred in the spring (12.4.1983) and the autumn (19.10.1983). The latter was much higher than the former. On the other hand, the highest peaks at Station B occurred in the autumn (19.10.1983) and the spring (20.3.1984). The highest peak found at this station was that of the spring. Biddulphia sinensis occurred at the same period during which the growth of B. paxillifer took place.

During the periods which preceded the growth of Biddulphia sinensis, the magnitude of variations of the environmental factors was very similar to that found prior to the high growth of B. paxillifer. In this case temperature varied from 5.5 to 18°C, nitrate from 5.7 to 66.73 µg at Nl^{-1}, nitrite from 0.15 to 0.63 µg at Nl^{-1}, ammonia from 0.05 to 0.39 µg at Nl^{-1}, silicate from 6.5 to 24.5 µg at Sil^{-1} and phosphate from 0.52 to 2.26 µg at Pl^{-1} (Table 5.2).

It was also found that Biddulphia sinensis and Bacillaria paxillifer usually dominate the phytoplankton populations together. See Figures 5.1.2, 5.1.4, 5.2.2 and 5.2.4. Therefore, it is possible that unmeasured environmental conditions may have stimulated the growth of B. sinensis or that different physiological races of B. sinensis were regulated by temperature over different temperature preferences (Vogelmann, 1980).

Table 5.2 Environmental factors measured during the growth of *Biddulphia sinensis*

FACTOR	STATION A		STATION B	
	4.3.83	6.9.83	6.9.83	17.2.84
Salinity (‰)	28.2	30.5	30.5	27
Temperature (°C)	5.5	18	18	5.5
Nitrate (μg at Nl^{-1})	33.57	5.8	5.7	66.7
Nitrite (μg at Nl^{-1})	0.2	0.6	0.63	0.15
Ammonia (μg at Nl^{-1})	0.21	0.34	0.39	0.05
Silicate (μg at Sil^{-1})	16.4	7.3	6.5	24.5
Phosphate (μg at Pl^{-1})	0.83	0.71	0.52	2.26

3. *Coscinodiscus sp.* (Figures 5.3.1 to 5.3.3)

Coscinodiscus sp. is the third in the group which includes the species which occur more or less all year round. The major occurrences of this species took place during the spring at both stations. An exceptional occurrence took place in the autumn (22.9.1983) at Station B (Fig. 5.3.1).

Table 5.3 shows the values of the environmental conditions which occurred prior to the onset of the *Coscinodiscus sp.* major growth. When it occurred in the spring, *Coscinodiscus sp.* was 20-50% dominant. The spring growth was observed over a temperature range of 5-11°C. The nutrient concentrations at that period were high. The autumn growth of *Coscinodiscus sp.* at Station B occurred in different conditions. At that period the nutrient concentrations were relatively low, and the temperature ranged from 18-19.5°C during the preceding months (July-

Table 5.3 Environmental factors measured during the growth of Coscinodiscus sp.

FACTOR	STATION A		STATION B		
	26.4.83	20.3.84	26.4.83	6.9.83	20.3.84
Salinity (‰)	26.7	30	26.6	30.5	30
Temperature (°C)	9	6.5	9	18	6.5
Nitrate (µg at Nl^{-1})	35.86	40.0	36.18	5.7	38.94
Nitrite (µg at Nl^{-1})	0.225	0.15	0.17	0.63	0.17
Ammonia (µg at Nl^{-1})	0.23	0.07	0.25	0.39	0.024
Silicate (µg at Sil^{-1})	17.05	15.38	16.17	6.53	15.06
Phosphate (µg at Pl^{-1})	1.1	1.28	0.99	0.52	1.41

September).

The autumn growth was 70% dominant. Once again, the growth of Coscinodiscus sp. was observed in two totally different conditions. A spring and autumn growth of Coscinodiscus sp. was observed. The wide range of environmental factors recorded give two possibilities to explain the results:

1. There were two populations of Coscinodiscus sp., a spring population adapted to cool water temperature and high nutrient concentrations and an autumn population which was adapted to high temperatures and low nutrient concentrations.
2. There was one population of Coscinodiscus sp.. The growth of this species was controlled by factors other than those recorded during the study.

B. A group of discontinuous occurrence

This group includes phytoplankton species which occur more than once but discontinuously.

1. Chaetoceros sp. (Figures 5.4.1 and 5.4.2)

In both stations, Chaetoceros sp. occurred mainly in the spring. The highest growth in 1983 occurred at the same time in both stations (11.5.1983), with the growth in the spring of 1984 characterized by a high bloom at Station B (14.5.1984) and a moderate growth in both stations in March of the same year.

In both stations, the salinity which preceded the increase was in the range 28.4-30.1‰. It can be observed from Table 5.4 that the increase of Chaetoceros on all the occasions followed a period of temperature in the range 7-11°C and high concentrations of nutrients. Chaetoceros sp. has been recorded over a temperature range of 9.8-19°C (Pearce, 1967; Paulraj, 1974; Vogelmann, 1980).

From the observations that Chaetoceros occurred over a wide temperature range, three distinct temperature preferences were identified (Vogelmann, 1980). The temperature ranges proposed are 10-13°C, 14-15°C and 16-19°C.

Based on this grouping the increase in Chaetoceros observed in this study (spring, 1983 and 1984) falls into the first temperature range. During this increase Chaetoceros sp. dominated the population at Station B (Fig. 5.4.2). On the other hand, the slight increase of Chaetoceros at Station A (11.7.1983) dominated the population reaching, about 43%. This slight increase during August occurred when the temperature was about 18°C.

Paulraj (1974) recorded a domination of Chaetoceros at Mumbles

Table 5.4 Environmental factors measured during the growth of Chaetoceros sp.

FACTOR	STATION A		STATION B		
	31.3.83	6.3.84	31.3.83	6.3.84	30.4.84
Salinity (‰)	28.6	29.1	28.4	29.4	30
Temperature (°C)	8	7	8	7	11
Nitrate (μg at Nl^{-1})	31.4	48.73	30.78	47.11	34.98
Nitrite (μg at Nl^{-1})	0.32	0.056	0.425	0.075	0.24
Ammonia (μg at Nl^{-1})	0.082	0.11	0.1	0.029	0.028
Silicate (μg at Sil^{-1})	13.39	17.41	13.7	16.95	8.38
Phosphate (μg at Pl^{-1})	0.9	1.35	0.81	1.034	0.92

during August, 1973 of 50-60%, when the temperature was 18-19°C. Vogelmann (1980) recorded domination of Chaetoceros during August, 1978 in Oxwich Bay at 18°C.

Vogelmann (1980) concluded from his results that the increase in Chaetoceros sp. in the summer could be stimulated by a factor other than the nutrients. The stimulating factor may have been the onset of a narrow temperature preference range.

In the present study, the highest increase of Chaetoceros occurred during the spring. At that time, nutrient levels were high enough not to limit the increase in the population. It appears that the increase from low winter to moderate spring temperatures, the increase in day length and hence the incidence of light, and the high level of nutrients all combined to stimulate the increase of Chaetoceros sp. during the spring. The high peak of Chaetoceros sp. at Station B

(14.5.1984, Fig. 5.4.1) may have been due to the spatial heterogeneity caused by the divergence in the sampling area (Collins et al., 1979). The increase of Chaetoceros sp. in August, 1983 (Station A) after a period of very low nutrient concentrations may be due to the narrow temperature preference range which has been suggested (Vogelmann, 1980)

2. Rhizosolenia hebetata (Figures 5.5.1 and 5.5.2)

In the present study, Rhizosolenia hebetata occurred mainly during the spring and early summer of 1983, 1984. The increase in the spring of 1983 was much higher than that of 1984 (Fig. 5.5.1).

R. Hebetata was found to occur over a wide range of temperatures, 7-14°C (Vogelmann, 1980). In the present study, the high increase of this species occurred at 8°C, 13.5°C and 18.5°C. In fact, the increase in numbers of this diatom followed periods of low and high temperatures. See Table 5.5. This means that it is not likely that tempera-

Table 5.5 Environmental factors measured during the growth of Rhizosolenia hebetata

FACTOR	STATION A		STATION B		
	4.3.83	17.2.84	4.3.83	23.6.83	17.2.84
Salinity (‰)	28.2	27	28.2	29	27
Temperature (°C)	5.5	5.5	5.5	15.5	5.5
Nitrate (µg at Nl^{-1})	33.57	65.99	32.54	6.69	66.73
Nitrite (µg at Nl^{-1})	0.2	0.18	0.187	0.237	0.15
Ammonia (µg at Nl^{-1})	0.21	0.042	0.37	0	0.046
Silicate (µg at Sil^{-1})	16.37	24.33	16.43	0.47	24.5
Phosphate (µg at Pl^{-1})	0.83	1.09	0.116	0.096	2.26

ture has controlled the occurrence of R. hebetata in Swansea Bay.

R. hebetata also occurred during a wide range of nutrient concentrations. The highest increase at Station A (18%) was preceded by high nutrient concentrations. On the other hand the highest increase of R. hebetata at Station B was preceded by very low nutrient concentrations. Hence, it can be concluded that nutrients did not appear to control the increase in R. hebetata. The only common factor which preceded the increase in R. hebetata was the salinity. Although R. hebetata is an oceanic diatom, its occurrence in Swansea Bay followed a period of relatively low salinity (27-29%o). This would suggest that the increase in R. hebetata was controlled by this range of salinity, but it must be noticed that R. hebetata has been observed along the south Gower coast over a relatively wide range of salinities (Vogelmann, 1980). Hence, it appears that the increase and succession of R. hebetata in Swansea Bay was controlled by unmeasured factor(s).

3. Melosira moniliformis (Figures 5.6.1 and 5.6.2)

This species occurred a few times during the present study. Its increase in the early summer (23.6.1983) in samples from both stations coincided with the highest spring-summer bloom of that year. In fact it increased to ca. 46% at Station A (58% dominant), and 9% at Station B (78% dominant).

The increase of M. moniliformis at both stations followed a period (March-May) of high nutrients and cool water temperatures (5.5-10°C). The only change in the measured factors was the increase in water temperature up to 13.5°C just before the bloom. See Table 5.6.

During and after the bloom there was a sudden decrease in the nutrients while the temperature remained high. It appears from these

Table 5.6 Environmental factors measured during the growth of *Melosira moniliformis*

FACTOR	STATION A 10.6.83	STATION B 10.6.83
Salinity (‰)	28	27.9
Temperature (°C)	13.5	13.5
Nitrate (μg at Nl^{-1})	35.86	36.55
Nitrite (μg at Nl^{-1})	0.375	0.39
Ammonia (μg at Nl^{-1})	0.07	0.06
Silicate (μg at Sil^{-1})	10.88	10.88
Phosphate (μg at Pl^{-1})	1.276	1.16

results that at the beginning, the increase in temperature triggered the increase of *M. moniliformis*. The exhaustion of the nutrients during that period may have caused the sudden drop in their numbers. At that time the nutrients became limiting, causing the termination of the *M. moniliformis* bloom.

4. *Prorocentrum micans* (Figures 5.7.1 to 5.7.3)

Prorocentrum micans was the main dinoflagellate which contributed largely to the total number of phytoplankton and dominated it at one time reaching 40%.

At both stations, *P. micans* occurred mainly during the summer and autumn months. See Figure 5.7.1.

In this study, *P. micans* was observed over a wide range of nutrient concentrations, from very high to very low concentrations.

This indicates that nutrients appear not to have controlled the increase of P. micans. As mentioned previously, P. micans occurred and increased during the summer and early autumn over a temperature range of 13-19°C. In fact, P. micans occurred only after the water temperature had reached over 11°C. See Table 5.7.

Table 5.7 Environmental factors measured during the growth of Prorocentrum micans

FACTOR	STATION A		STATION B	
	25.5.83	13.6.84	25.5.83	13.6.84
Salinity (‰)	28	30	27.7	30.2
Temperature (°C)	11.5	14	11.5	14
Nitrate (μg at Nl^{-1})	37.88	5.25	38.07	5.37
Nitrite (μg at Nl^{-1})	0.19	0.075	0.26	0.075
Ammonia (μg at Nl^{-1})	0.044	0.12	0.031	0.145
Silicate (μg at Sil^{-1})	14.21	1.927	13.82	1.862
Phosphate (μg at Pl^{-1})	1.2	0.116	0.773	0.174

It appears that temperature was the limiting factor of P. micans growth as is the case with most of the dinoflagellates (Gran and Braarud, 1935; Tait, 1981). It can also be concluded that P. micans was adapted to the low nutrient concentrations during the onset of its favourable range of temperature. That may have been the reason for its dominance (40%) of the phytoplankton population when the nutrient concentrations were very low.

5. Rhizosolenia shrubsolei (Figures 5.8.1 and 5.8.2)

R. shrubsolei occurred in the summer months of 1983 and 1984. Its major occurrence was in July, 1983. It increased up to 9.5% at Station A and 6% at Station B. R. shrubsolei followed the bloom of M. moniliformis in a period of very low levels of nutrients during the summer of 1983 (Table 5.8).

Table 5.8 Environmental factors measured during the growth of Rhizosolenia shrubsolei

FACTOR	STATION A 23.6.83	STATION B 23.6.83
Salinity (‰)	28.8	29
Temperature (°C)	15.5	15.5
Nitrate (µg at Nl^{-1})	6.85	6.69
Nitrite (µg at Nl^{-1})	0.218	0.237
Ammonia (µg at Nl^{-1})	0.0135	0
Silicate (µg at Sil^{-1})	0.977	0.474
Phosphate (µg at Pl^{-1})	0.086	0.096

In both years it was observed over a narrow range of temperatures (14-18°C) and it appears that this factor was critical to the appearance of the species in the plankton. But during its occurrence, R. shrubsolei dominated the phytoplankton community (60%, A and 45%, B).

6. Skeletonema costatum (Figures 5.9.1 and 5.9.2)

Skeletonema costatum is a major representative of the temperate water phytoplankton (Lebour, 1929). This species has been observed in the Bristol Channel in the spring over a temperature range of 6-11°C (Pearce, 1967; Paulraj, 1974; Tyler, 1976; Vogelmann, 1980). Although S. costatum has been observed in most of the studies carried out along the south Gower coast, in most of the offshore studies (Tyler, 1976; Vogelmann, 1980) it was not found to be more than 40% dominant.

In the present study, the increase in S. costatum followed a period of cool water temperature (Table 5.9) and when it was found, the water temperature was 11°C (30.4.1984) which is within the suggested preference range.

Table 5.9 Environmental factors measured during the growth of Skeletonema costatum

FACTOR	STATION A 20.3.84	STATION B 20.3.84
Salinity (‰)	30	30
Temperature (°C)	6.5	6.5
Nitrate (μg at Nl^{-1})	40.03	38.94
Nitrite (μg at Nl^{-1})	0.15	0.17
Ammonia (μg at Nl^{-1})	0.073	0.024
Silicate (μg at Sil^{-1})	15.38	15.06
Phosphate (μg at Pl^{-1})	1.27	1.41

In the present study, S. costatum blooms at Stations A and B were 28% and 38% dominant respectively (Fig. 5.9.2). From the results it appears that in Swansea Bay, S. costatum occurred when the nutrient concentrations were high enough and the temperature was within the preference range.

7. Thalassionema nitzschioides (Figures 5.10.1 and 5.10.2)

Thalassionema nitzschioides has not been a familiar member of the phytoplankton community along the south Gower coast, but it occurred a few times during the present study, reaching 52% dominance at one time (Fig. 5.10.2). From the results it appears that T. nitzschioides was one of the few phytoplankton species which survived the extreme conditions of the cold months. For example there was a negligible number of cells of this species in December, 1982 (Fig. 5.10.1), but it comprised 33% of that month's phytoplanktons.

C. Species with one major occurrence

Although the species included in this group occurred on one occasion only, they represented a significant percentage of the highest total phytoplankton population encountered.

1. Rhizosolenia delicatula (Figures 5.11.1 and 5.11.2)

R. delicatula was observed in June, 1984. It increased to 96% (Station A) and 10% (Station B) and it was 95% and 71% dominant, respectively (Figs. 5.11.1 and 5.11.2). It occurred during and after a period of very low nutrient concentrations. This was accompanied by a narrow range of warm temperature (14-15°C) and relatively high salinity (30-31‰) (Table 5.10). It is possible that these restricted conditions were favourable for R. delicatula to increase and compete successfully.

Table 5.10 Environmental factors measured during the growth of Rhizosolenia delicatula

FACTOR	STATION A 13.6.84	STATION B 13.6.84
Salinity (‰)	30	30.2
Temperature (°C)	14	14
Nitrate (μg at Nl^{-1})	5.255	5.37
Nitrite (μg at Nl^{-1})	0.075	0.075
Ammonia (μg at Nl^{-1})	0.121	0.145
Silicate (μg at Sil^{-1})	1.927	1.862
Phosphate (μg at Pl^{-1})	0.116	0.174

2. Thalassiosira gravida (Figures 5.12.1 and 5.12.2)

Thalassiosira gravida was observed during the spring of 1984 only (14.5.1984). See Figure 5.12.1. The maximum increase was in May (4.5%, A and 17%, B).

This species was dominant (22%, A and 41%, B) after a decline in the nutrient concentrations and when the water temperature was 11°C (Table 5.11).

Although the measured environmental factors were similar during the growth period, there is a difference of magnitude between the growth of T. gravida at Station A and that at Station B. This difference in growth may have been the result of the spatial heterogeneity in the area of sampling.

It can be seen from the results in Table 5.11 and Figures 5.12.1 and 5.12.2 that T. gravida increased in number during the periods of

low silicate and phosphate concentrations. This indicates that this species may have been a successful competitor within the phytoplankton community at low nutrient concentrations, but growth itself was stimulated by the onset of a favourable water temperature.

Table 5.11 Environmental factors measured during the growth of Thalassiosira gravida

FACTOR	STATION A		STATION B	
	30.4.84	14.5.84	30.4.84	14.5.84
Salinity (‰)	30.1	31	30	30.7
Temperature (°C)	11	11.5	11	11.5
Nitrate (µg at Nl^{-1})	34.62	19.02	34.98	19.02
Nitrite (µg at Nl^{-1})	0.225	0.237	0.244	0.225
Ammonia (µg at Nl^{-1})	0.058	0.008	0.028	0
Silicate (µg at Sil^{-1})	8.04	1.0	8.38	0.85
Phosphate (µg at Pl^{-1})	1.0	0.232	0.918	0.247

SUMMARY

Phytoplankton group and species succession in Swansea Bay followed a more or less regular pattern. Diatoms occurred at different times of the year at temperatures lower than those at which the dinoflagellates occurred. Some of the species performed a regular and expected occurrence such as Biddulphia sinensis, Bacillaria paxillifer, and Prorocentrum micans (Figs. 5.13 and 5.14). On the other hand some species occurred at times when the conditions were apparently more specific. Due to one reason or another, a few species occurred in high numbers only once, e.g. Thalassiosira gravida.

In both stations, B. paxillifer and B. sinesis occurred in high numbers and dominated the phytoplanktons mainly during the autumn. Chaetoceros sp., Rhizosolenia sp., and M. moniliformis dominated the spring and early summer populations. Dinoflagellates, e.g. P. micans dominated the late summer and early autumn populations (Figs. 5.13 and 5.14).

In the present study, some differences in the species composition and the degree of dominance were observed between Stations A and B.

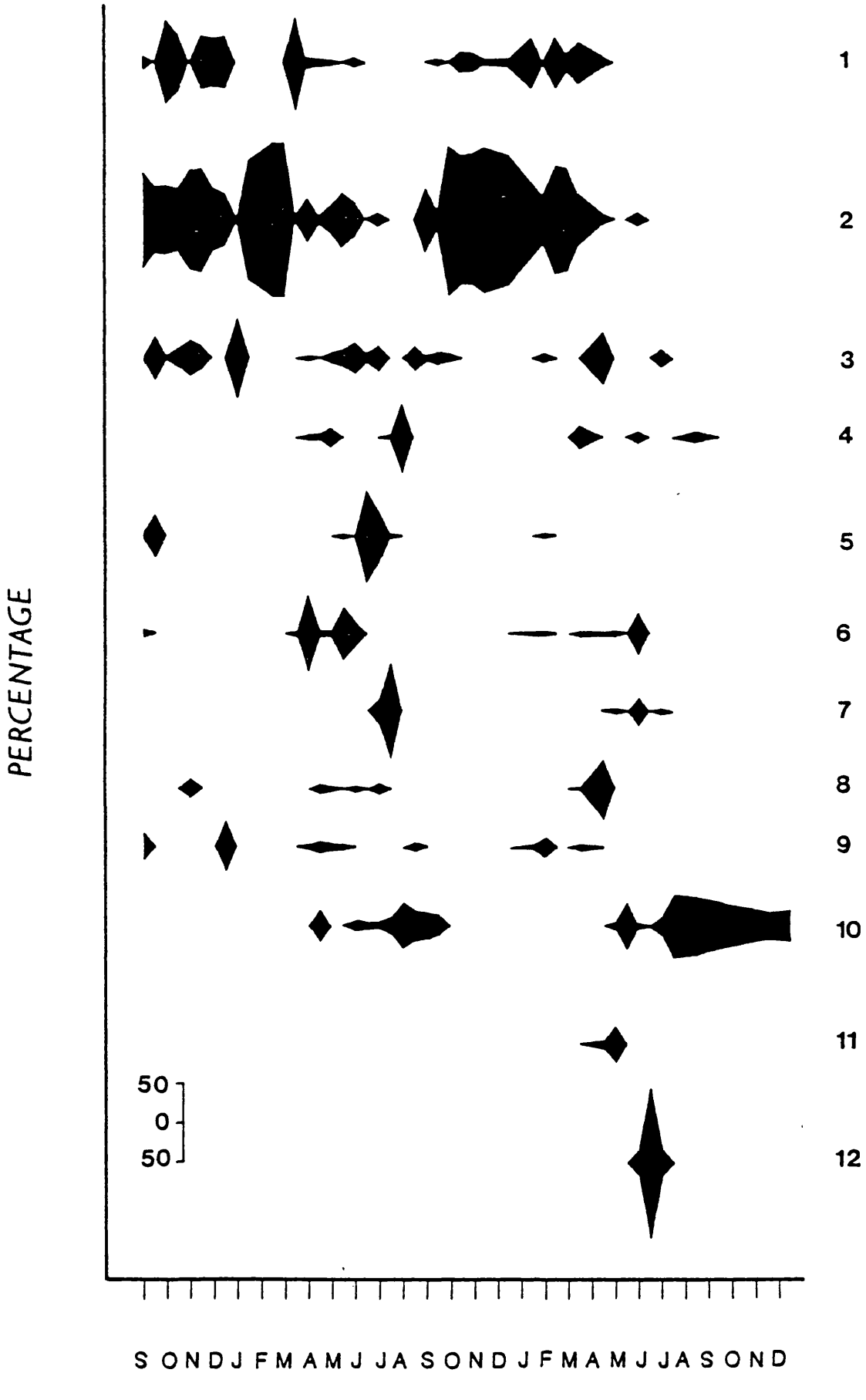
For example on 21.9.1982, Coscinodiscus sp. represent 68% of the phytoplankton of that sample at Station B, while it represented 28% only at Station A. On 22.8.1983, Chaetoceros sp. represented 43% of the populations of Station A, while it only represented 14% at Station B. Another example is the sample of 14.5.1984. At Station B, Thalassiosira gravida represented 42% of the populations, while in the other station it represented 22%.

The chemical factors which were recorded on the dates mentioned previously were very similar at both stations. This leaves the hydro-

Figure 5.13 Phytoplankton dominance in Swansea Bay (Station A)

1. Bacillaria paxillifer
2. Biddulphia sinensis
3. Coscinodiscus sp.
4. Chaetoceros sp.
5. Melosira moniliformis
6. Rhizosolenia hebetata
7. Rhizosolenia shrubsolei
8. Skeletonema costatum
9. Thalassionema nitzschioides
10. Prorocentrum micans
11. Thalassiosira gravida
12. Rhizosolenia delicatula

A


PERCENTAGE
1
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10
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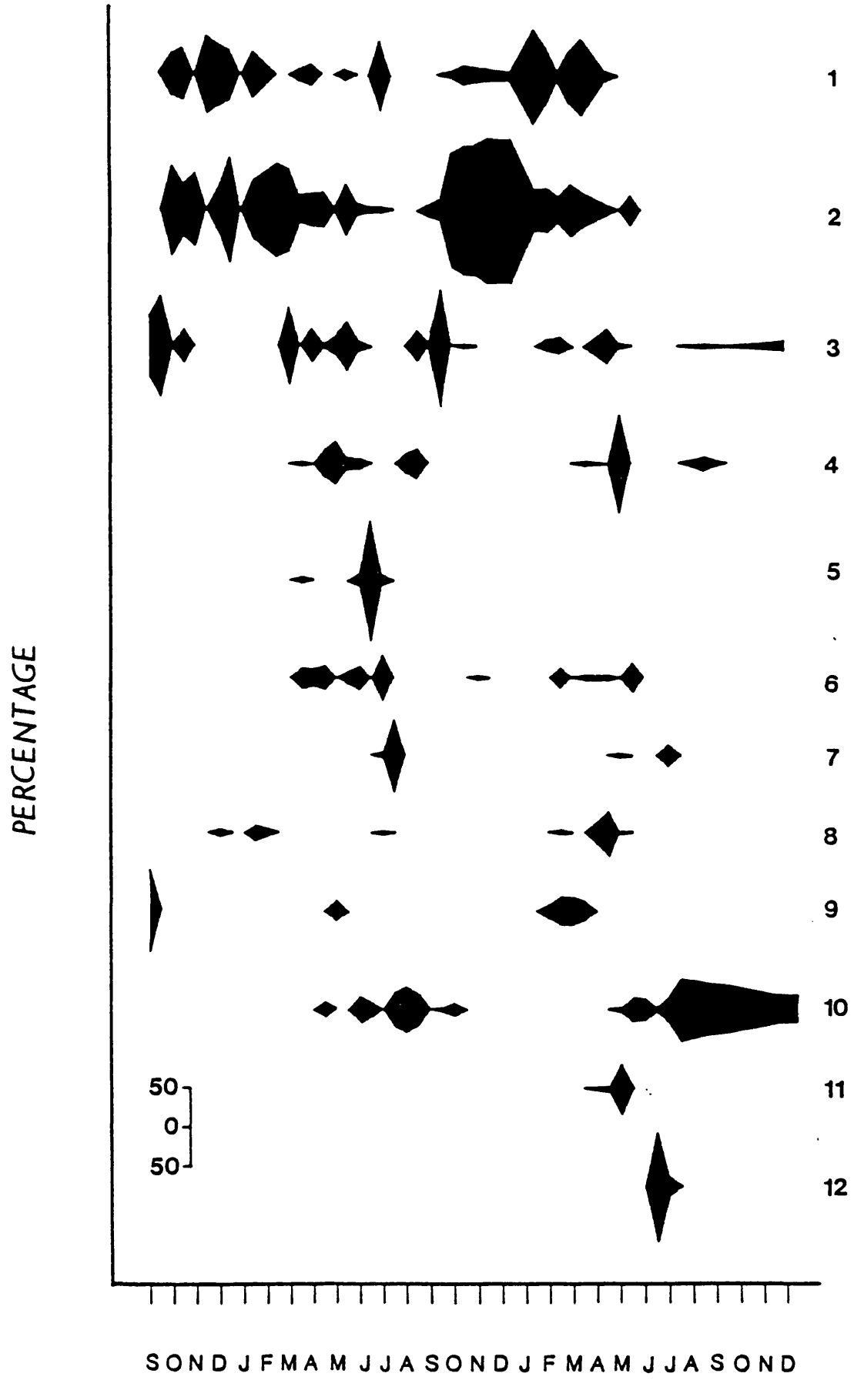
50
0
50
S O N D J F M A M J J A S O N D J F M A M J J A S O N D
1982
1983
1984

Figure 5.14 Phytoplankton dominance in Swansea Bay (Station B)

1. Bacillaria paxillifer
2. Biddulphia sinensis
3. Coscinodiscus sp.
4. Chaetoceros sp.
5. Melosira moniliformis
6. Rhizosolenia hebetata
7. Rhizosolenia shrubsolei
8. Skeletonema costatum
9. Thalassionema nitzschioides
10. Prorocentrum micans
11. Thalassiosira gravida
12. Rhizosolenia delicatula

B


PERCENTAGE
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SONDJFMAMJJASONDJFMAMJJASOND
1982
1983
1984

graphic conditions as the reason for the differences encountered at the two stations. An area of divergence has been thought to occur in the area of sampling (Collins et al., 1979). This proposed divergence may have been the main factor which produced the differences in the phytoplankton of Stations A and B. Another possibility is the transport of phytoplankton populations from other parts of the bay as a result of the eddy circulation (Collins et al, 1979; Joint, 1980). This point is further discussed in the general discussion chapter.

CHAPTER VI

PRIMARY PRODUCTIVITY OF PHYTOPLANKTON SIZE FRACTIONS

INTRODUCTION

The role of phytoplankton as primary producers has been known for a long time. For most of the years spent in studying the production of phytoplankton, net plankton have always been regarded as the major (if not the only) contributors to the production of aquatic environment.

This approach has been derived from reliance on net samples for assessments of population composition and biomass. The term nanoplankton was first used by Lohmann (1903), referring to the part of plankton not retained by the net. It was then that the possible contribution of nanoplankton to the biomass and production was suggested. Since Lohmann (1903), many definitions have been suggested to describe nanoplankton.

In the present study, the term nanoplankton will be used to describe the phytoplankton cells between 2-20 μm (Dussart, 1965; Sieburth et al., 1978).

Sieburth et al. (1978) gave the name picoplankton to the small plankton which are in the size range 0.2-2.0 μm. Based on Lohmann's (1903) definition of nanoplankton, their importance in the phytoplankton has been proved by many studies (Steemann Nielsen, 1938; Atkins, 1945; Harvey, 1950; Knight-Jones and Walne, 1951; Wood and Davies, 1956; Malone, 1971a; Durbin et al., 1975; Throndsen, 1978; Hannah and Boney, 1983). The contribution of nanoplankton to the total phytoplankton productivity has been estimated from different localities: Steemann Nielsen and Jensen (1957) 82%, Yentsch and Ryther (1959) 98%, Teixeira (1963) 90%, Malone (1971a) 60-90%, Durbin et al. (1975) 51%, Hannah and Boney (1983) 50%.

On the other hand, the contribution of picoplankton to the biomass

and production of phytoplankton has only recently been investigated (Sieburth et al., 1978). Since then data have accumulated giving more evidence of the importance of picoplankton (0.2-2.0 µm).

From recent studies, more evidence supports the findings that there is considerable production by photoautotrophic organisms which are of the same size as bacteria. The major contribution to the production is no longer restricted to the net plankton and the nanoplankton >5 µm. A series of recent findings in different localities has shown that nanoplankton < 5 µm and picoplankton (0.2-2.0 µm) are very important quantitatively. In the size fraction <1 µm, production was found to represent 20 to 30% of the total (Gieskes et al., 1979); 20 to 80% (Li et al., 1983); 60% (Platt et al., 1983).

Joint and Pomroy (1983) reported primary production values of 35 to 40% by organisms >1 - <5 µm and 20 to 30% by organisms < 1 µm. Organisms which passed through a 3 m filter and representing up to 25% of the phytoplankton biomass have been reported (Larsson and Hagström, 1982).

In general, nano- and picoplankton are more abundant and productive than net plankton especially in the offshore and oceanic waters (Steemann Nielsen and Jensen, 1957; McAllister et al., 1959; Malone, 1971b; Semina, 1972; Johnson and Sieburth, 1979; Waterbury et al., 1979; Li et al., 1983; Platt et al., 1983). On the other hand, net plankton increases in abundance towards continental shelf and coastal waters (Hulbert, 1962, 1970; Strickland et al., 1969; Ryther et al., 1971; Malone, 1971a, 1976, 1977a).

Several exceptions to this general pattern have been observed in shallow, temperate estuaries and adjacent coastal waters influenced by estuarine runoff (Ryther, 1954; Loftus et al., 1972; Durbin et al.,

1975; Malone, 1976, 1977b; Hannah and Boney, 1983).

EXPERIMENTAL

From each station, 3 x 100 ml seawater samples were poured into 125 ml bottles, two clear and one dark. 2 µci $NaH^{14}CO_3$ were added to each bottle at zero time. After 4 hours of incubation, 7 x 10 ml portions from each bottle were filtered through 0.45, 1, 5, 10, 20, 50 and 80 µm filters. The filtration, treatment of filters, and counting of radioactivity were carried out in the usual way (Chapter II).

RESULTS

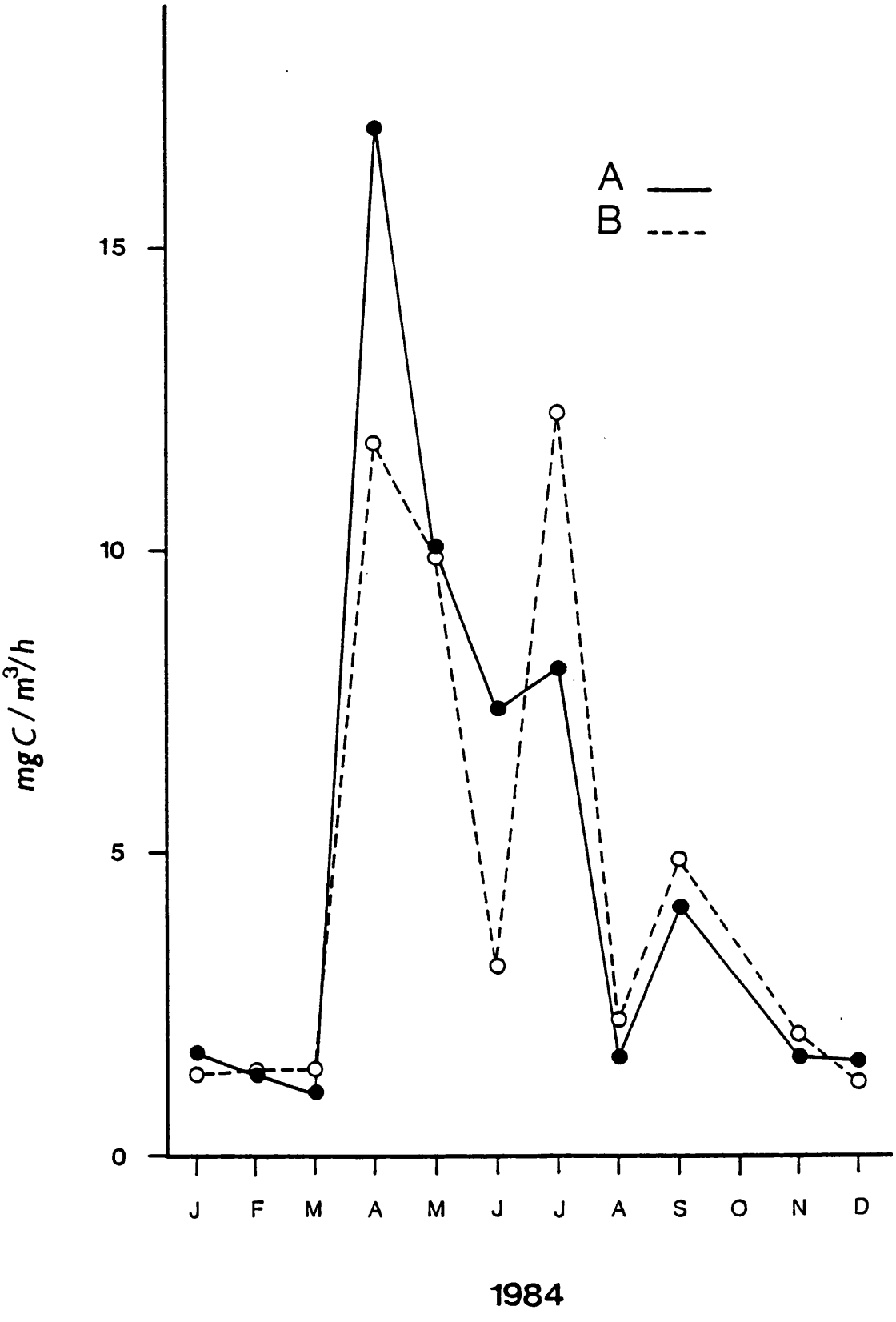
The results of one year of sampling are treated in two different ways. To determine the significance of each size class in relation to the monthly total, the productivity of the size class was calculated as a percentage of the total productivity of that month. On the other hand, the seasonal variation of each size class was calculated based on the highest productivity value found in the whole year as 100%.

A. Total primary productivity

In this one year long programme, the primary productivity at each station was determined monthly. At both stations the highest values occurred in the spring-summer and in the autumn. The highest value of primary productivity (ca. 17 mg cm^{-3} hr^{-1} was found at Station A in April (Fig. 6.1).

Figure 6.1 Total primary productivity

A: eastern station
B: western station


A
B
15
10
5
0
mg C / m3/h
J
F
M
A
M
J
J
A
S
O
N
D
1984

B. Monthly percentage of each size class

These are the values of each size class based on the total primary productivity of each month as 100%.

1. 0.45-1 µm

Station A: Monthly average = 14.28%

The values here were variable throughout the year (Fig. 6.2). The values were in the range of 0.3-37.4%. The highest values were found in March (31.3%), May (37.4%) and December (33%).

Station B: Monthly average = 19.16%

The values of this size class at this station were different from Station A. The values were in the range of 2.2-43.4% (Fig. 6.2). The period of highest production extended from March to May. At this period the highest production was in March (43.4%). The highest value in the period starting in the summer and ending in the winter occurred in August (30.8%).

2. 1-5 µm

Station A: Monthly average = 22.35%

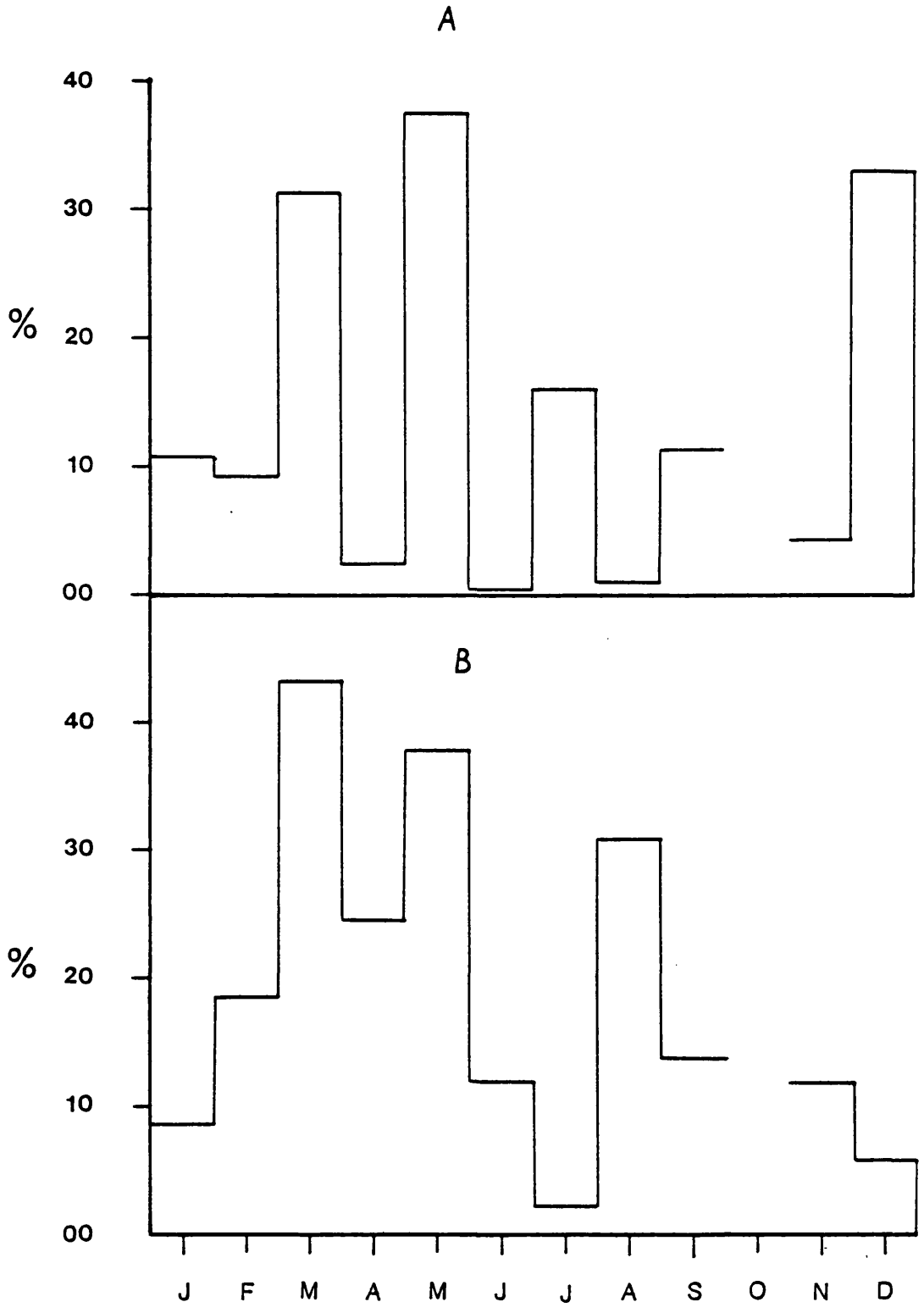
At this station, the productivity of autotrophic organisms >1 - <5 µm was very important. They were highly productive from March to September. In that period there was little variation in the values of primary productivity. The values ranged from 17.1% in July to the highest value of 50% in March with an average of 30.6% (Fig. 6.3).

Station B: Monthly average = 11.58%

In contrast to Station A, there was a period extending from January to

Figure 6.2 Monthly percentage of 0.45 - 1 μm size class

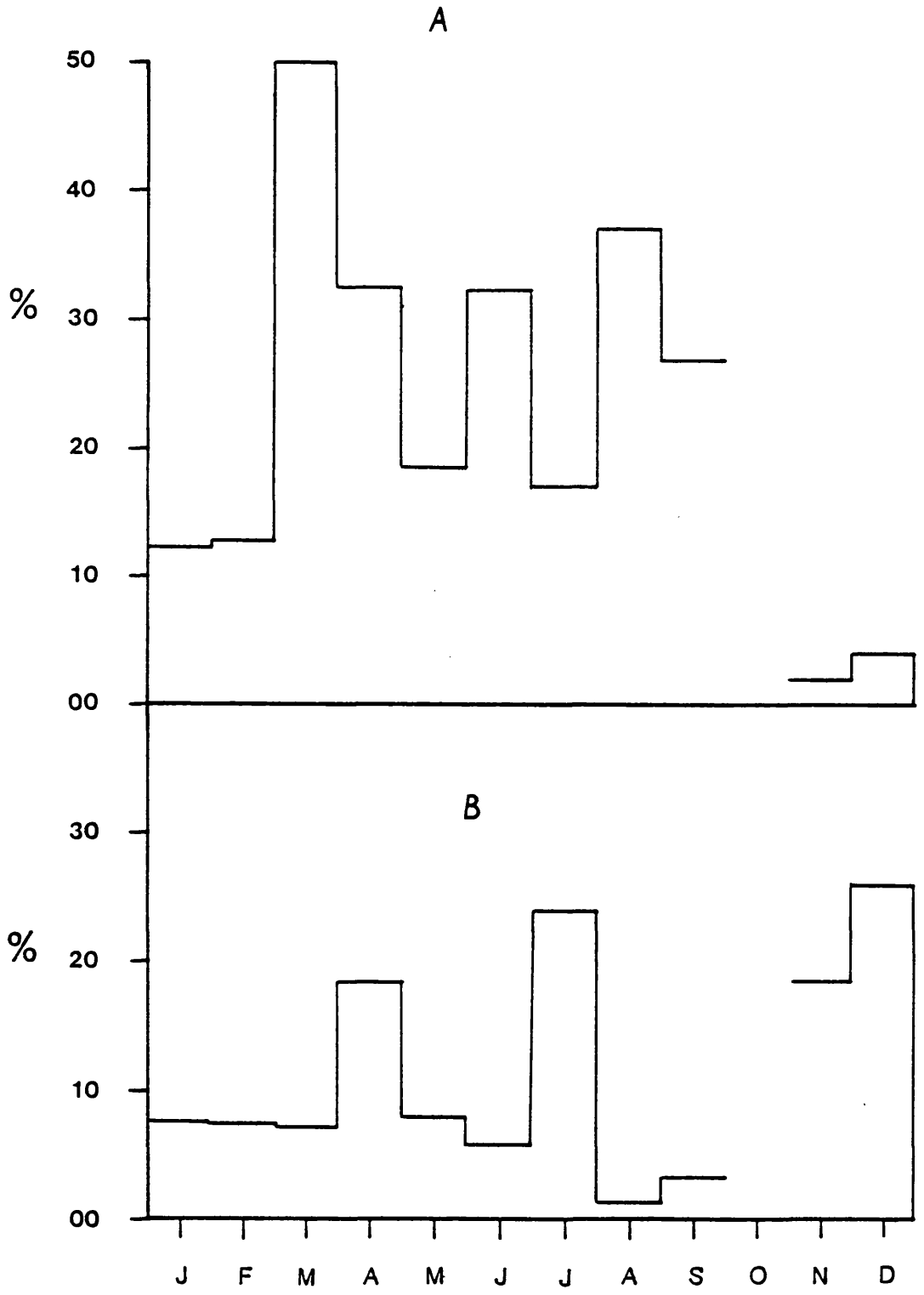
0.45-1 μm



1984

Figure 6.3 Monthly percentage of 1 - 5 μm size class

1 - 5 µm



1984

September with fairly constant productivity value ca. 5.8%. There were exceptional values to the average of that period. Those values were 18.5% (April) and 23.9% (July). In this station, the values of November and December were different from those of Station A with values of 18.5% and 26.1% respectively (Fig. 6.3).

3. 5-10 µm

Station A: Monthly average = 24.67%

In this size class the highest values have occurred mainly from May to September and they were over a range of 21.7% (August) to 39.5% (September). The highest production value in this size class (56.5%) occurred in November (Fig. 6.4).

Station B: Monthly average = 26.48%

As in Station A, the highest values of Station B occurred mainly from May to September and they were over a range of 26.2% (July) to 65.4% (August). The lowest values were found in the winter months and early spring with an average value of 8.35% (Fig. 6.4).

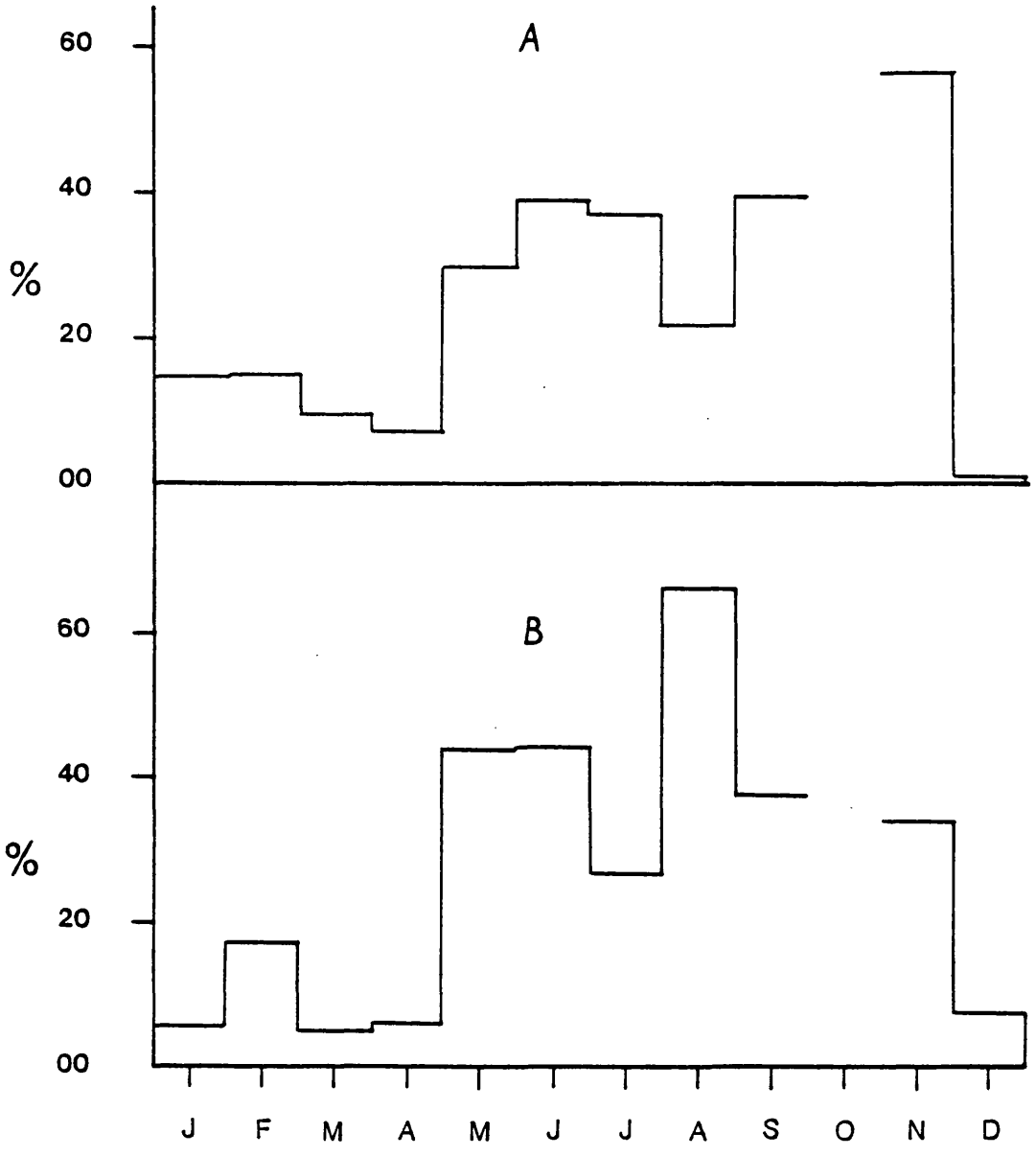
4. 10-20 µm

Station A: Monthly average = 18.56%

In this size class, the lowest percentage was 7.6 (May), and the highest was 35.4 (January). The high production values in this size class were not restricted to one season or one continuous period but distributed almost evenly throughout the year (Fig. 6.5). The four highest values occurred in January (35.4%), February (26.5%), April (31.8%) and August (25.6%).

Figure 6.4 Monthly percentage of 5 - 10 μm size class

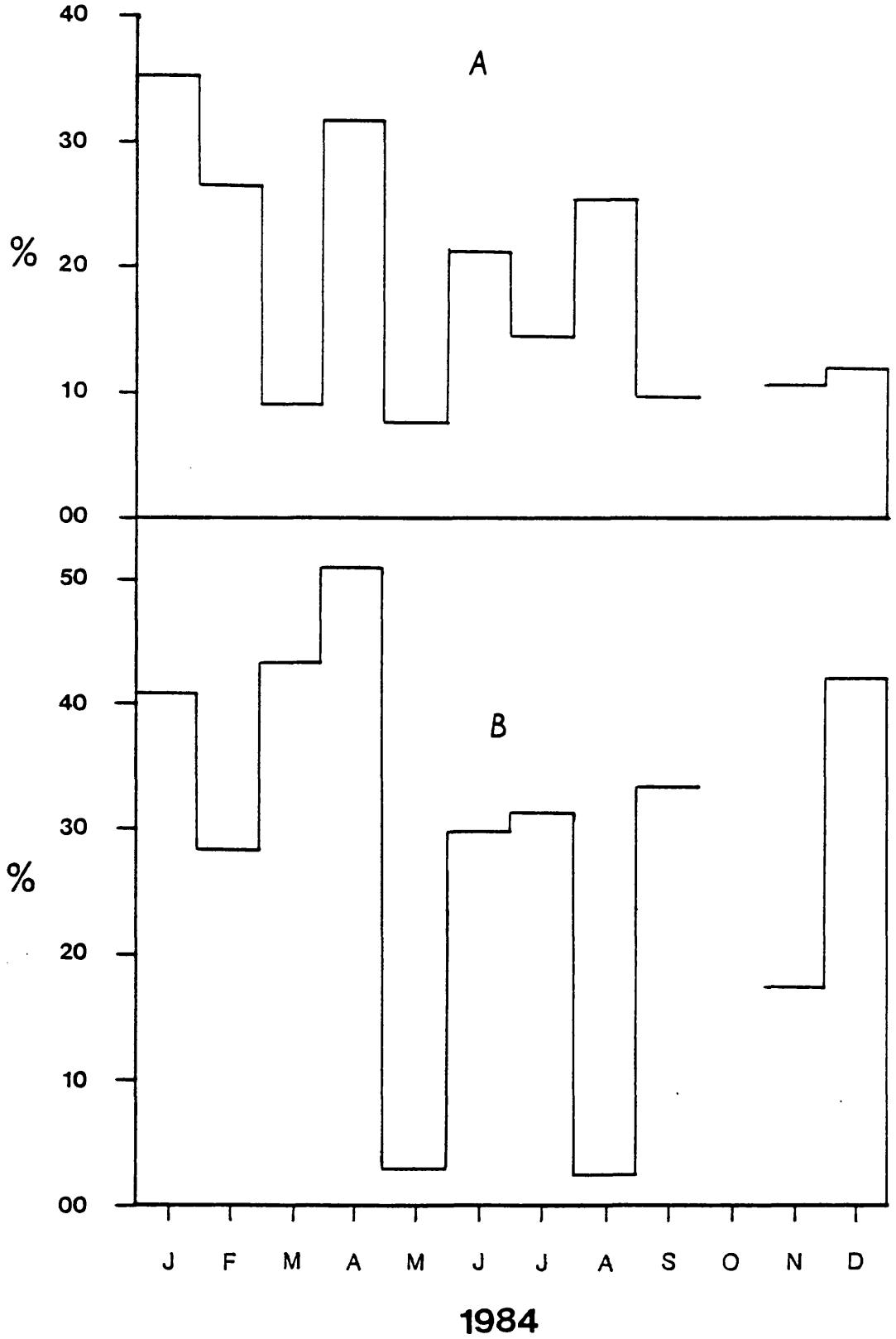
5 - 10 μm



1984

Figure 6.5 Monthly percentage of 10 - 20 µm size class

10 - 20 μm



Station B: Monthly average = 29.37%

The organisms represented in this size class are important and regular contributors to the primary productivity of phytoplankton. This can be shown clearly by excluding the values of May and August from the total of this class. The remaining values ranged from 17.4% (November) to 43% (April) with an average of 35.28% (Fig. 6.5).

5. 20-50 µm

Station A: Monthly average = 15.29%

The highest contribution to the total primary production by the phytoplankton of this size class took place mainly in the winter, January (22%), February (16.4%), November (21.6%) and December (37.94%).

In addition to that, an average contribution of 11.7% took place in the spring and summer (from April to September). See Figure 6.6.

Station B: Monthly average = 10.5%

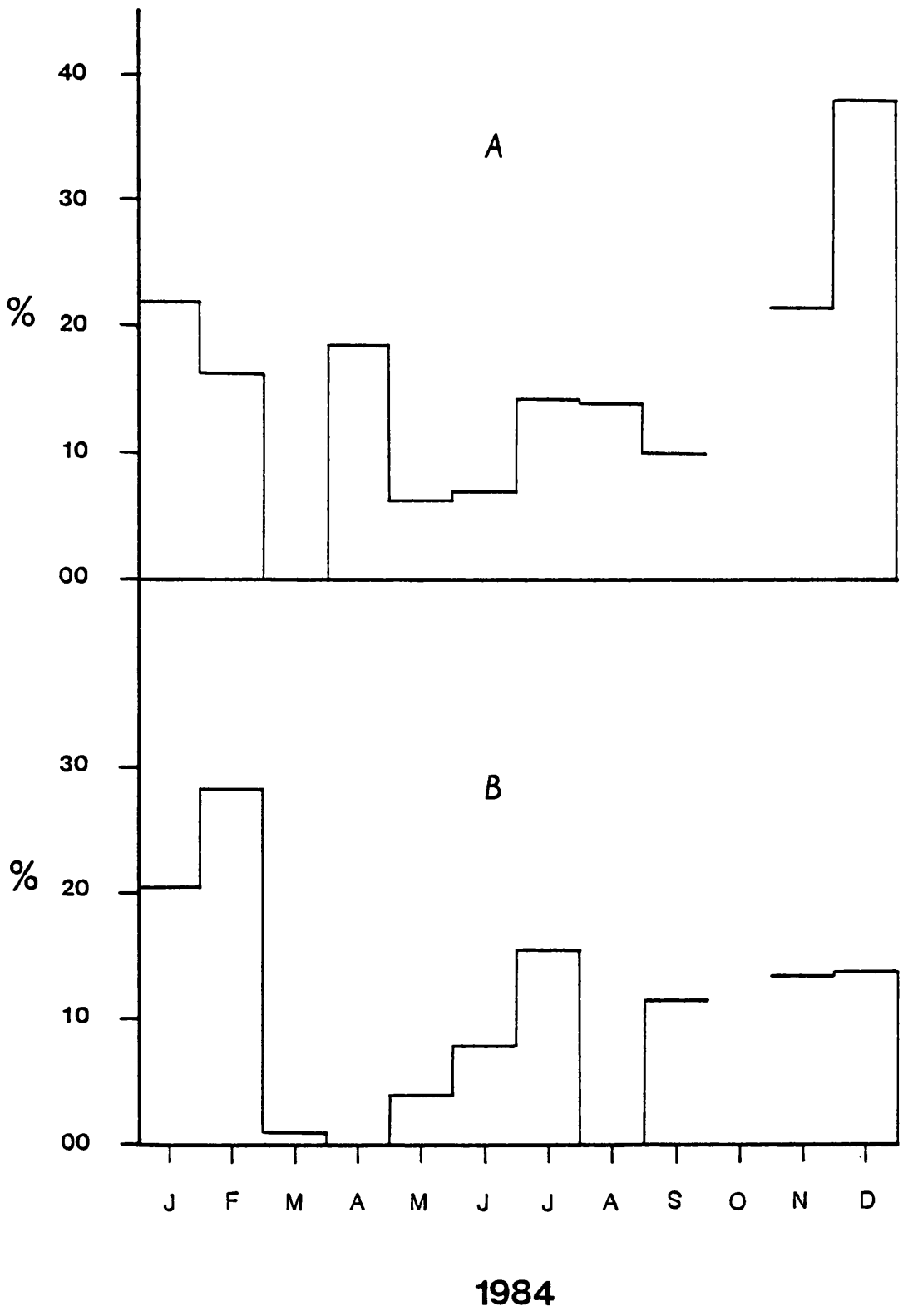
As in Station A, the highest contribution to the total production by the organisms in this size class occurred in the winter, January (20.4%), February (28.4%), November (13.4%) and December (13.64%). The average monthly contribution of May, June and July was ca. 9% (Fig. 6.6).

6. 50-80 µm

Station A: Monthly average = 4.8%

The organisms of this size class were not very important contributors to the total primary production of each month. The highest contributions took place in February (19.3%) and December (12%). During the rest of the year the production was very low (Fig. 6.7).

Figure 6.6 Monthly percentage of 20 - 50 μm size class


20 - 50 µm
40
30
%
20
10
00
A
30
%
20
10
00
B
J
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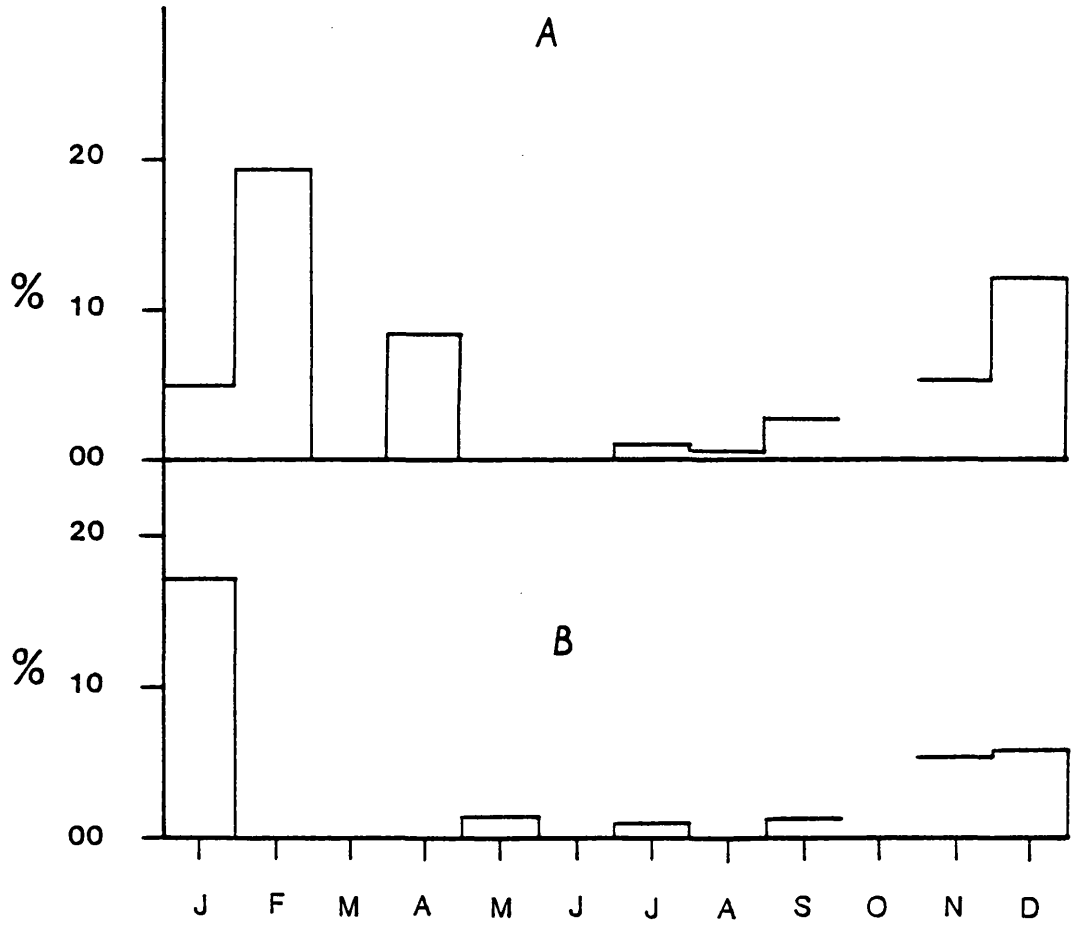
M
J
J
A
S
O
N
D
1984

Figure 6.7 Monthly percentage of 50 - 80 μm size class

50 - 80 µm



1984

Station B: Monthly average = 2.9%

As in Station A, the contribution to the monthly total primary production by the organisms of this size class was very low. The highest value occurred in January (17.2%), while the monthly average of the remaining months of the year was 1.5% (Fig. 6.7).

C. Seasonal variation of phytoplankton size classes

These are the values of the monthly production by the different size classes based on the highest total production (16.99 mg cm^{-3} hr^{-1}) as 100%.

1. 0.45-1 µm

Station A:

The values of primary production in this size class were usually less than 3%. Two exceptions were found, the values of May (21.9%) and July (7.42%). See Figure 6.8.

Station B:

As in Station A, the primary productivity values at Station B were usually low. April and May were characterized by high values (14% and 22.74% respectively). See Figure 6.9.

2. 1-5 µm

Station A:

Here the period of high values extended from April to September over a range of 3.41-30.47%. This period was characterized by a noticeable high value of production in April (30.47%). See Figure 6.8.

Figure 6.8 Seasonal variation of the size classes:
(Station A)

0.45 - 1 µm
1 - 5 µm
5 - 10 µm

A

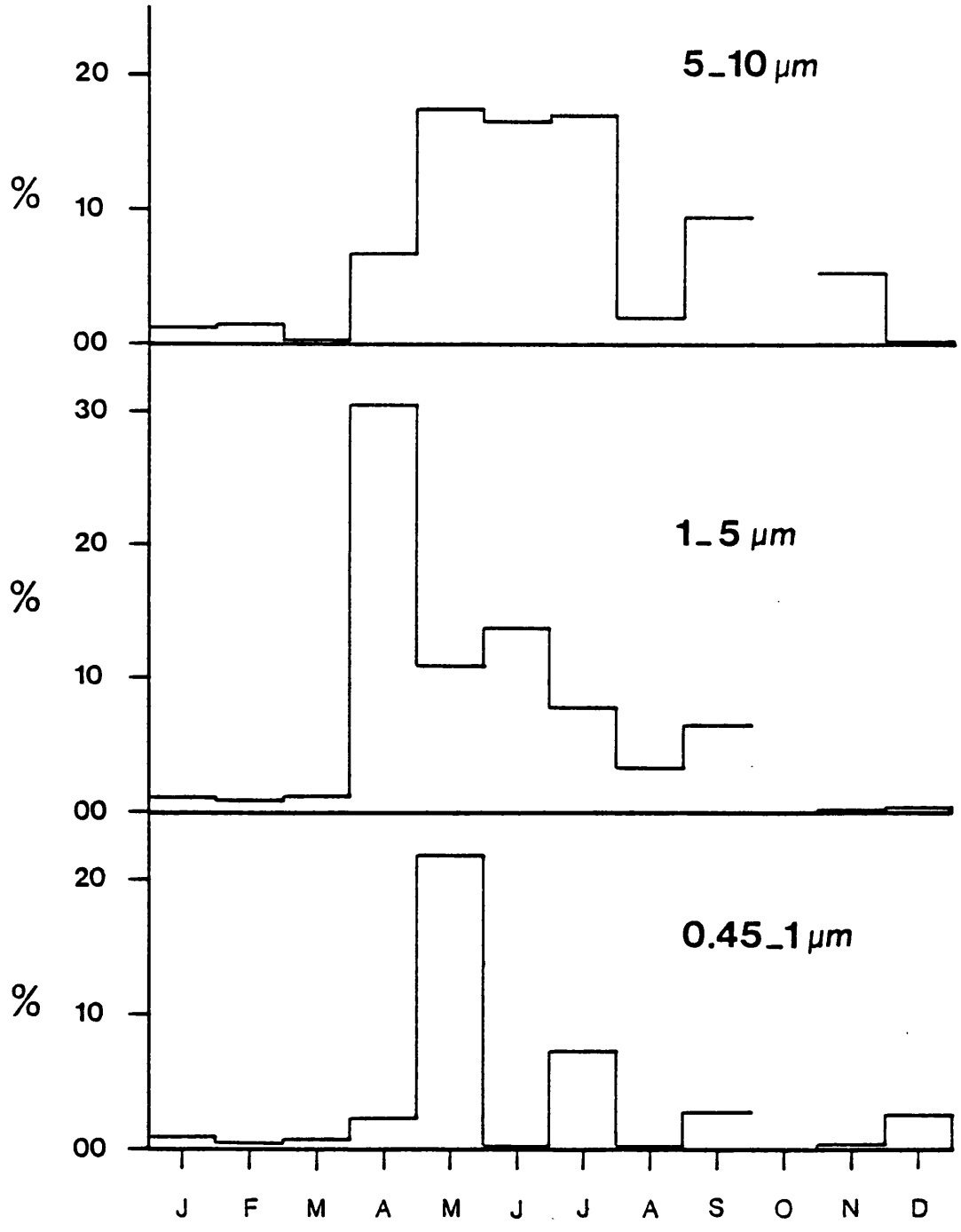
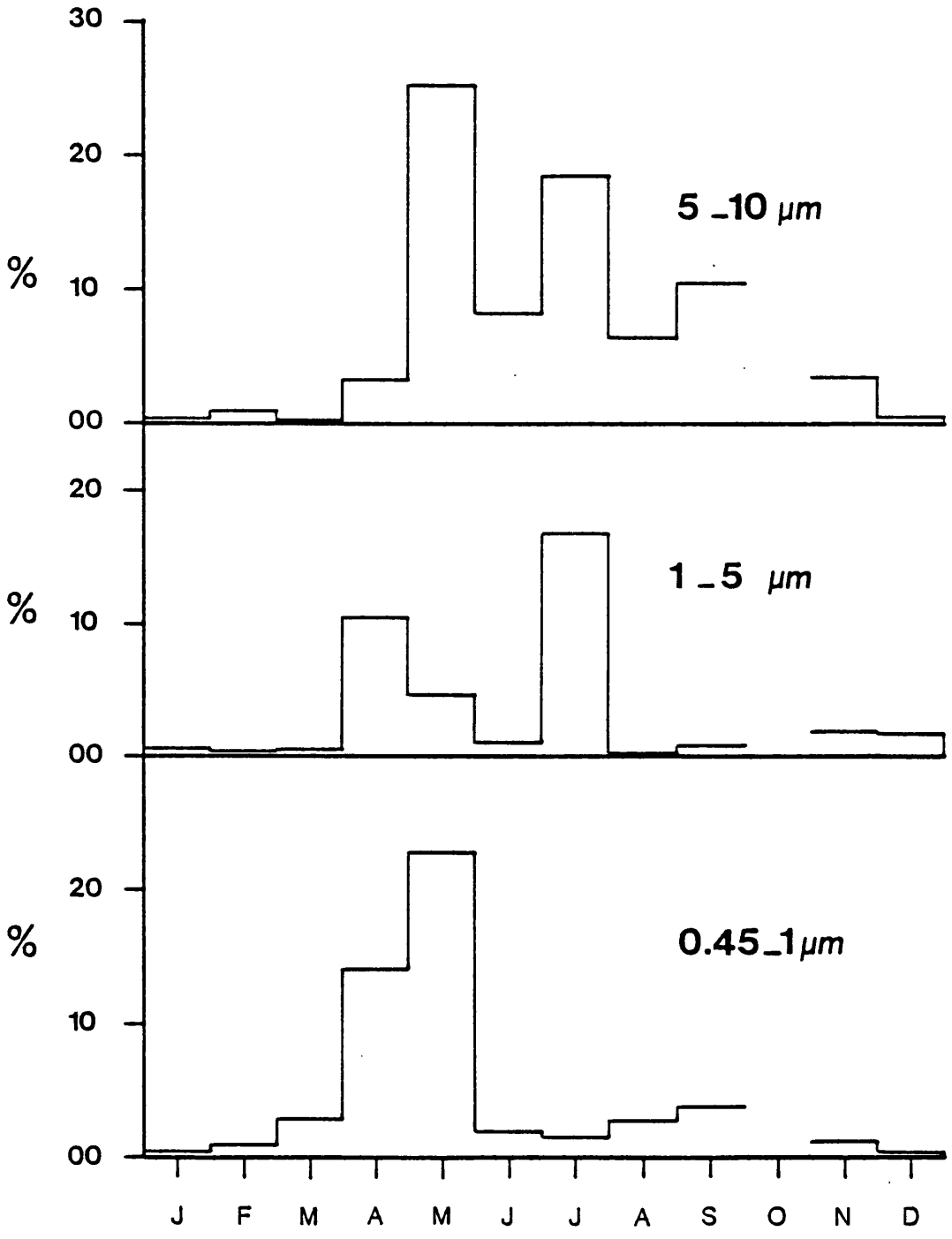


Figure 6.9 Seasonal variation of the size classes:
(Station B)

0.45 - 1 µm
1 - 5 µm
5 - 10 µm

B


30
20
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5 _10 µm
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10
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%
1 _5 µm
20
10
00
%
0.45_1µm
J F M A M J J A S O N D
1984

Station B:

The distribution of the significant values at this station was different from Station A. The high values of production extended from April to July (Fig. 6.9). In this period of time the values ranged from 1.06% in June to 16.83% in July (Fig. 6.9).

3. 5-10 µm

Station A:

At this station, the distribution of production values in this size class was characterized by a period of high and constant production extending from May to July. The average of the production values in this period was 17.1% (Fig. 6.8).

Station B:

The pattern of distribution in this station was to a certain extent similar to that at Station A. The period of high production extended from May to September. The values of production in this period were more variable than those at Station A. They were over a range of 6.06-25.19% with an average value of 13.66% (Fig. 6.9).

4. 10-20 µm

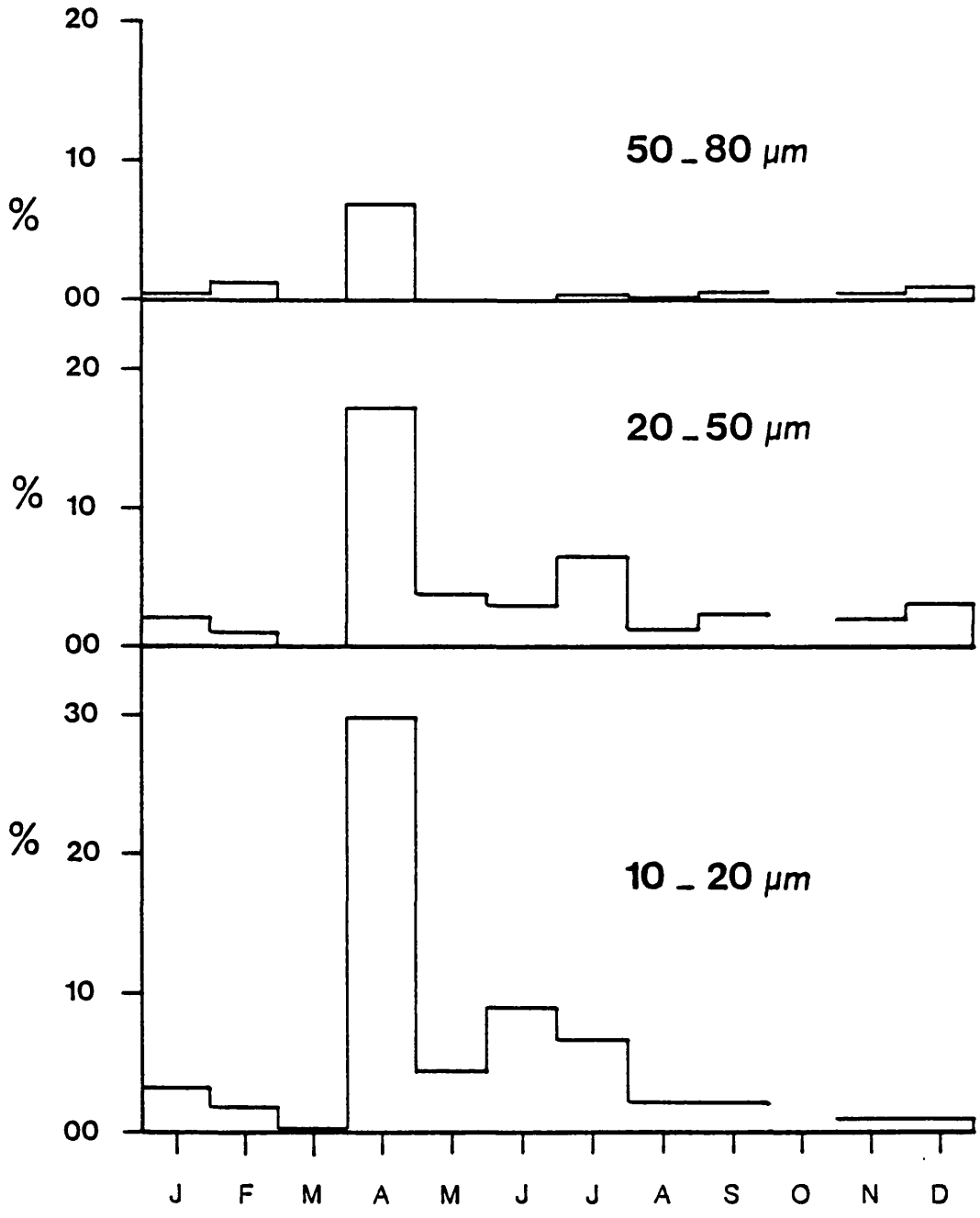
Station A:

The phytoplankton in this size class contributed to the production throughout the year. On the other hand, this contribution was usually low except for the period from April to July (Fig. 6.10). The production values in this period were around 7%. The highest contribution by this size class was in April (29.82%).

Figure 6.10 Seasonal variation of the size classes:
(Station A)

10 - 20 µm
20 - 50 µm
50 - 80 µm

A


20
10
%
00
50 _ 80 µm
20
%
10
00
20 _ 50 µm
30
%
20
10
00
10 _ 20 µm
J F M A M J J A S O N D
1984

Station B:

At this station, the production values were variable throughout the year (Fig. 6.11). They were generally low except on two occasions when the production was 29.25% (April) and 22.13% (July).

5. 20-50 μm

Station A:

Throughout the year, production values were relatively low (Fig. 6.10). The highest production values occurred during the spring and early summer. The highest value in that period was found in April (17.38%).

Station B:

As in Station A, the values here were low most of the time (Fig. 6.11). The highest value was found in July (10.95%).

6. 50-80 μm

Station A:

The production values were very low throughout the year (<1%) except for one occasion (April) when it reached 6.93% (Fig. 6.10).

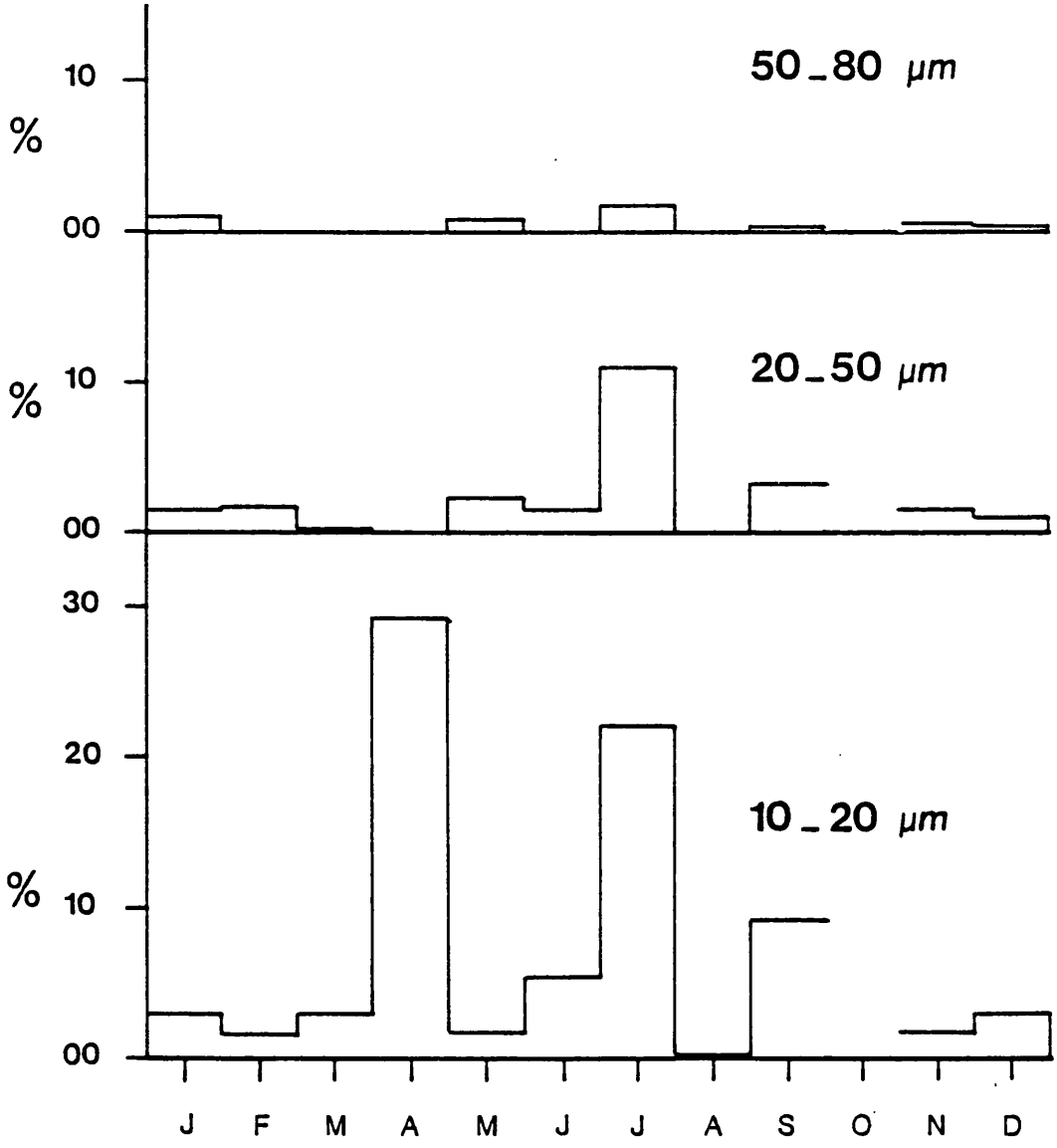
Station B:

At this station, the production values were very low throughout the year (<1.71%). See Figure 6.11.

Figure 6.11 Seasonal variation of the size classes:
(Station B)

10 - 20 µm
20 - 50 µm
50 - 80 µm

B



DISCUSSION

The fractionation technique used in this study was based on the assumption that the different size classes of phytoplankton can be separated by the filtration of seawater through filters of different pore size.

It has been reported that nucleopore filters behave as screens (Sheldon, 1972), but they may still retain particles smaller than the rated pore size. Joint and Pomroy (1983) tested the retention of bacteria on 5 and 1 µm pore size filters by counting the number of bacteria in a water sample before and after their standard fractionation procedure and found recoveries of between 90.3 and 99.8%. Nucleopore filters have been used in the present study because of their satisfactory reliability as separators of different particle sizes. Very low vacuum was used during the filtration procedures in order to minimize possible cell fragmentation.

Filtration of the water sample into different size classes was carried out after incubation with ^{14}C. This was carried out in order to avoid damage to the phytoplankton by the filtration before they take up the ^{14}C. The data obtained in this study are presented in two ways:

A. Monthly percentage

This approach shows the significance of each size class to the monthly total production.

Station A: (Figures 6.2 to 6.7)

From the figures it can be seen that the winter populations of phytoplankton were dominated by phytoplankton 10-50 µm in size which were

mainly diatoms. By early spring, phytoplankton of the size class 0.45-5 μm began to establish themselves and appeared as the major contributors to production. During the spring and summer months most of the production was carried out by phytoplankton of the size class 0.45-10 μm. This was during the time when small diatoms (<20 μm) and flagellates appeared to be the dominant groups (Chapter V). This high contribution to the monthly production continued until late autumn.

Station B: (Figures 6.2 to 6.7)

At this station, the highest monthly production by organisms > 20 μm occurred during the winter. The spring production was carried out by the phytoplankton size class of 0.45-20 μm. It seems that although the net plankton appeared dominant and major contributors to the monthly production during the spring, the picoplankton (0.45-1 μm) were very important. In fact they contributed up to 43% of the production. By late spring and early summer the role of organisms 10-20 μm began to decrease and organisms 5-10 μm started to become the major contributors to the monthly production.

From the Figures 6.2 to 6.7 it can be seen and concluded that plankton of the size 0.45-20 μm were very important to the primary production in Swansea Bay. If the total production of organisms 0.45-20 μm was calculated as a percentage of the total monthly primary production it represented 50-100% (A) and 62.3-100% (B).

B. Seasonal variation

This approach shows the significance of each size class throughout the year.

Station A: (Figures 6.8 and 6.10)

These figures show clearly that when the seasonal significance of the different size classes was considered, the most important size classes were 1-5 and 5-10 μm. They were highly productive during the spring-summer months. When the values of 1-5 and 5-10 μm size classes (April-July) were considered as one unit, they represented 24.9-37.22% of the highest annual production.

In addition to the role of phytoplankton of the size class 1-10 μm, organisms which passed 1 μm pore size nucleopore filter appeared to make a significant contribution occasionally. Organisms < 1 μm represented 21.9% of the total primary production in May.

The larger members of the phytoplankton were absent at certain periods and contributed nothing to the ^{14}C fixation (Fig. 6.10). Organisms from 10-80 μm did not contribute significantly to the production except on one occasion. This contribution occurred in April which is quite understandable because of the increase in net plankton during early spring.

Station B:

As in Station A, the major contribution of organisms <1 μm occurred mainly in May. On the other hand, the highest productivity was achieved by organisms in the size classes 1-5 and 5-10 μm. When the April-July values of these two size classes were added together, they represented 9.24-35.31%.

The organisms in the size class 10-20 μm had a more significant role at this station than those of Station A. During the April to July period they occurred twice as significant contributors to the primary production, once in April (29.25%) and the other in July (22.13%).

Organisms of the size classes 20-50 µm and 50-80 µm were not significant contributors to primary production.

From the previous review it is interesting to see that in Swansea Bay the nano- and picoplankton have a far more important role than would have been expected.

In general, pico- and nanoplankton play a very important role in the oceanic environments (Semina, 1972; Waterbury et al., 1979; Platt et al., 1983). The idea of nanoplankton dominance in the oceanic environment only has been subject to increasing exceptions (Loftus et al., 1972; Durbin et al., 1975; Malone, 1976; Hannah and Boney, 1983). The possible significant role of nanoplankton in Swansea Bay was first suggested by Paulraj and Hayward (1980). The results obtained in the present study are similar to those of Bruno et al. (1983), and Hannah and Boney (1983). Although in the study of Hannah and Boney nanoplankton were generally dominant in the winter, water surface data show that nanoplankton were much more significant in the spring and summer than in winter. Bruno et al. (1983) working on primary productivity and phytoplankton size fraction dominance in a temperate North Atlantic estuary found that nanoplankton (<20 µm) accounted for 88.5% of the productivity and 88.1% of the standing crop during May through September. On the other hand, net plankton (>20 µm) has been found to account for higher primary productivity percentage than nanoplankton during the winter months.

From the results of the present study it can be seen that the highest rate of productivity occurred during spring and summer. Most of that period was characterized by having high monthly percentages of pico- and nanoplankton productions.

The high pico- and nanoplankton production took place during a

Table 6.1 Nutrient concentrations during the fractionation programme (μg at l^{-1})

A

DATE	NITRATE (N)	NITRITE (N)	AMMONIA (N)	SILICATE (Si)	PHOSPHATE (P)
24. 1.84	26.7	0.15	0.11	16.4	0.58
17. 2.84	66.0	0.18	0.042	24.3	1.8
20. 3.84	40.0	0.15	0.073	15.39	1.28
30. 4.84	34.6	0.225	0.058	8.04	1.0
31. 5.84	0.2	0.019	0.048	3.33	0.15
27. 6.84	2.81	0.094	0.027	0.49	0.15
11. 7.84	0.6	0.056	0.037	0	0.17
30. 8.84	5.94	0.92	0.3	5.16	0.52
13.11.84	31.4	0.63	0.066	16.5	1.18
11.12.84	34.8	0.225	0.058	13.4	1.45

B

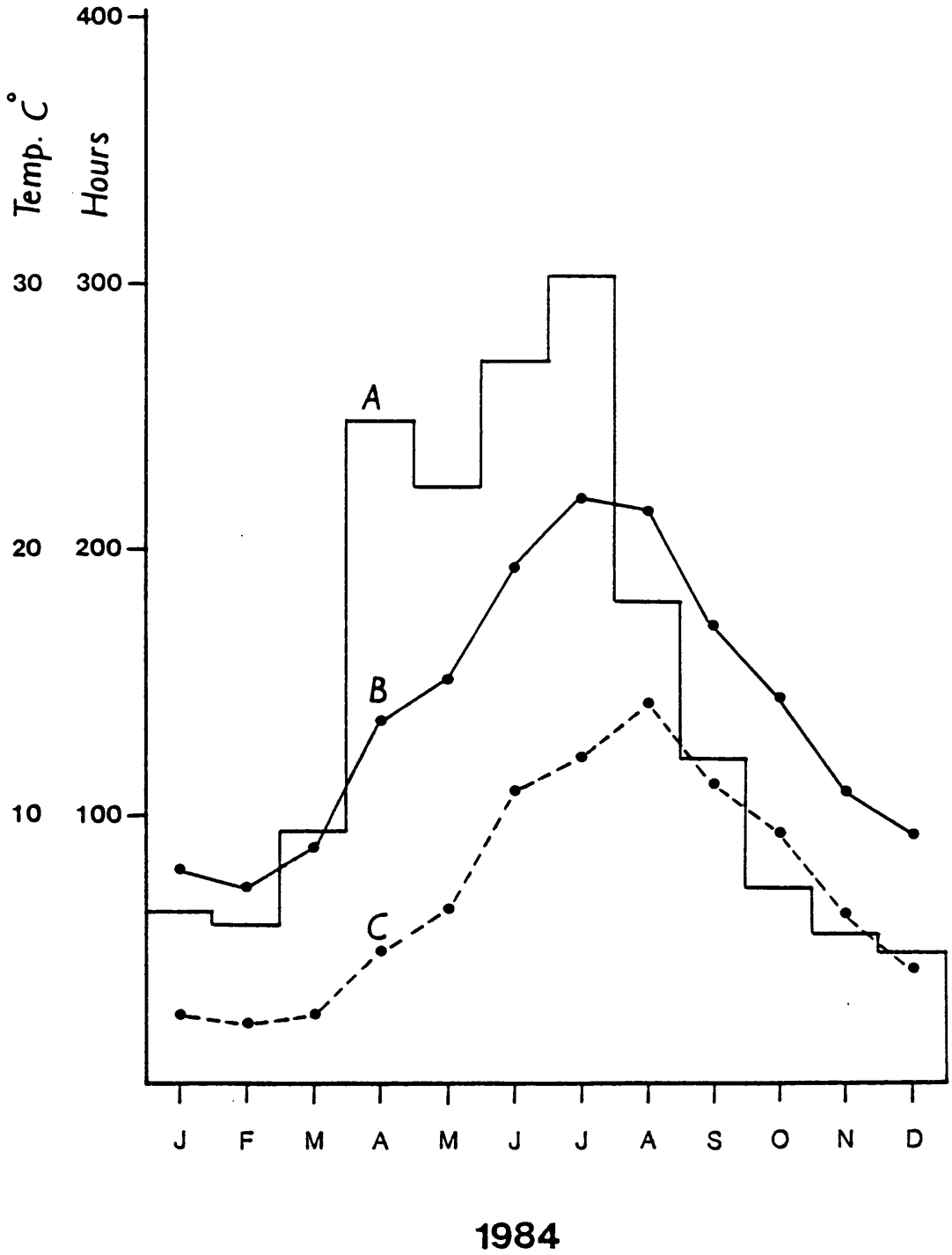
DATE	NITRATE (N)	NITRITE (N)	AMMONIA (N)	SILICATE (Si)	PHOSPHATE (P)
24. 1.84	26.0	0.13	0.05	15.1	0.95
17. 2.84	66.7	0.15	0.046	24.5	2.26
20. 3.84	38.9	0.17	0.024	15.06	1.41
30. 4.84	35.0	0.24	0.028	8.4	0.92
31. 5.84	0.14	0.019	0.042	1.73	0.1
27. 6.84	1.72	0.14	0	0.85	0.06
11. 7.84	0.82	0.056	0	0	0.21
30. 8.84	4.4	0.54	0.365	4.12	0.37
13.11.84	31.52	0.62	0.063	16.53	1.24
11.12.84	35.65	0.29	0.03	13.13	1.57

period of low nutrient concentrations (Table 6.1). And because these organisms did not represent high percentage during winter months when nutrient concentrations were at the highest level, it can be said that high nutrient concentrations were not controlling pico- and nano-plankton production. On the other hand, the spring-summer period is characterized by higher temperature and longer sunshine periods. The availability of high nutrient concentrations (Table 6.1) and the increasing daily sunshine hours (Fig. 6.12) may have favoured the increase in net plankton during the winter-early spring period. This is because the lower the surface area/volume ratio, the lower the ability to uptake more nutrients (Eppley et al., 1969). These conditions may have favoured the higher production percentage of net plankton with low surface area/volume ratio in the winter and early spring.

The increase in pico- and nanoplankton production (Figs. 6.8 and 6.9) occurred in a period of high temperature, high monthly sunshine hours, and low nutrient concentrations. As mentioned previously, net plankton (>20 μm) were more significant when the high sunshine hours were accompanied by high nutrient concentrations. Considering the advantages nanoplankton have in terms of surface area/volume ratio, and therefore nutrient uptake (Eppley et al., 1969) and presumably growth rate (Williams, 1964), as a result they have outgrown net plankton during periods of low nutrient concentrations.

Figure 6.12 Monthly sunshine hours and air temperature

A: Total monthly sunshine hours
B: Maximum air temperature
C: Minimum air temperature


400
300
200
100
Temp. C°
Hours
30
20
10
A
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C
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CHAPTER VII

LABORATORY EXPERIMENTS

INTRODUCTION

Since the early years of marine biology, growing algae in the laboratory has been one of the main tasks facing scientists. Natural waters were used as growth media in algal cultures, but they have been found unsatisfactory mainly because some essential nutrients are usually present in only trace amounts, their concentrations depending on dynamic equilibria which are disturbed as soon as the water is collected (Fogg, 1975). Natural waters supplemented with various nutrients have been much used when the object has been only to produce algal material, precise knowledge of the conditions affecting its growth being unnecessary. One of the earliest artificial media having some resemblance to those in which algae grow naturally was devised by Chu (1942). Thereafter, many improvements of artificial media have been introduced and many new artificial media have been prepared (Rhode, 1948; Krauss, 1953; Miller and Fogg, 1957; Provasoli et al., 1957; Droop, 1961; Stein, 1973; Morel et al., 1979). Algal cultures were developed to a degree that they can be used to explain the interaction between phytoplankton and its surrounding environment. Two different types of cultures are used for this purpose, batch culture and continuous culture. Batch culture is the simplest and most commonly employed culture system. Algal cells are inoculated into a nutrient solution contained in a suitable vessel which is often shaken or aerated to suspend the cells, replenish CO_2 and otherwise maintain a homogeneous environment. Growth in batch culture exhibits a very distinctive curve. The establishment of an exponential phase at the early growth period makes batch culture a suitable technique to study natural populations with a similar growth phase e.g. Spring bloom

(Droop, 1975; Myklestad, 1977). Batch cultures are extremely useful in autoecological studies as long as the environment and possible cell-cycle effects are taken into account. They have been used most successfully to study the effect of parameters such as light intensity, temperature and salinity on growth rates and to provide material for countless studies on ultrastructure and chemical composition. Continuous culture is a relatively new kind of culture developed in the 1940s. This type of culture was developed to overcome the problem of very high cell concentrations in batch culture and to study the physiological status of phytoplankton under steady-state growth rate and nutrient limitation. Continuous cultures were used to study nutrient limitation because a hyperbolic relationship between the cellular fraction of the limiting nutrient and the growth rate has been found (Fuhs, 1969; Caperon and Meyer, 1972; Passche, 1973; Droop, 1974; Tilman and Kilham, 1976). Nutrient limitation is believed to be the most important factor which controls phytoplankton competition and species succession. Nutrient limitation is based on the competition theory which predicts that, under idealized conditions (Titman, 1976), the one species best able to acquire and use the limiting source (Liebig, 1843) should displace all other competing species. In the natural environment this situation has never been found. It is always the case that many phytoplankton species co-exist in the same body of water. This observation has been termed the paradox of the plankton (Hutchinson, 1961). One of the explanations for this contradiction between theory and the real situation is that species-specific, nutrient-utilization characteristics result in different species each being limited simultaneously by a different nutrient. Direct competition is avoided and potentially as many species can co-exist as

there are limiting nutrients (Tilman, 1977). Other explanations stress that the mixed layer of lakes and oceans is not homogeneous as is generally assumed. Well-known vertical gradients in light, nutrients and temperature during calm weather provide spatial heterogeneity, allowing localized increases in those populations which find themselves in favourable conditions (Darley, 1982). Quantification of the ability to take up nutrients, given optimal levels of all other growth factors has been attempted (Kuenzler and Ketchum, 1962; Fuhs, 1969; Carpenter, 1970). Significant differences have been observed in the nutrient uptake capacities of different species, and this finding has often been suggested as a possible controlling factor in natural phytoplankton species succession.

Redfield (1958) reported that the C:N:P atomic ratio of particulate matter in sea water is 106:16:1 and that these nutrients appear to be depleted in a similar proportion to this ratio during phytoplankton growth. When the utilization of N and P was closely examined in surface water, N was found to be depleted first with a significant amount of P always remaining in solution (Ryther and Dunstan, 1971). Therefore, N is generally considered as the limiting nutrient in sea water.

Using the Redfield ratio alone as an indicator to nutrient limitation may be misleading. This is because whereas it is usually safe to assume that nutrient concentrations above the µg at l^{-1} range (e.g. 1 µg at Pl^{-1}) are not limiting (Darley, 1982), lower, even very low concentrations may or may not be limiting depending on the presence of other limiting factors and the rate at which the nutrient is recycled in the system (Rhee, 1978). Therefore, additional indicators should be used to support the possibility of nutrient limitation due

to the low concentration of a certain nutrient. Nutrient-enrichment bio-assay is the technique most commonly used to investigate nutrient limitation in algal cultures and natural communities. Care must be taken in the interpretation of the result obtained as productivity in the enrichment experiments. The result obtained shortly after enrichment will be misleading because the productivity values will not be representative of the real response of phytoplankton to nutrient addition. Nutrient-limited phytoplankton will be using the available energy to assimilate nutrient rather than take up additional carbon (Gerhart and Likens, 1975; Thomas et al., 1976; Hipkin et al., 1983).

In the present study, special consideration was given to the effect of nitrate on the growth of, and ^{14}C-fixation by, algae in artificial media and natural sea water. This is because low nitrate values have been found at certain times of the year. When these values were compared with phosphate values found at these times, low N to P ratios (< 16) were obtained. The possible nitrate limitation to the growth of phytoplankton was further investigated using a bio-assay technique.

Materials and Methods

I. Culture media and phytoplankton growth

i. Growth media:

a. Erdschreiber medium:

Erdschreiber medium was used mainly for maintaining the phytoplankton species. This medium was made up as follows:

$NaNO_3$ stock solution	1 ml
$Na_2HPO_4.12H_2O$ stock solution	1 ml
Soil extract*	50 ml
Vitamin B-12 solution	1 ml
Vitamin B-1 solution	1 ml
Fe/EDTA solution	1 ml
Trace element solution	1 ml
Fluorosilicate solution	1 ml

Made up to 1 litre with filtered sea water

The stock solutions were prepared as follows:

$NaNO_3$ stock solution	20 g/100 ml
Na_2HPO_4 stock solution	3 g/100 ml
Cyanocobalamin (B-12) solution	0.1 g/250 ml; diluted tenfold for stock solution
Thiamine (B-1) solution	0.1 g/250 ml; diluted tenfold for stock solution
Fe/EDTA solution	1 ml $FeCl_3$ solution and 0.23 g Na_2 EDTA/100 ml
Fluorosilicate solution	0.1 g sodium fluorosilicate/100 ml
Soil extract	1 kg of garden soil added to 2 litres of distilled water, autoclaved for 1 hour at 5 lbs pressure, cooled and centrifuged * soil extract was filtered before use through Whatmann No.1 filter papers
Trace element solution	0.1 g $ZnSO_4.7H_2O$ 0.001 g $CoCl_2.6H_2O$ 0.002 g $MnCl_2.4H_2O$ 0.001 g $Na_2MoO_4.2H_2O$ 0.001 g H_3BO_3 0.0002 g $CuSO_4.5H_2O$ in 100 ml double distilled water

All solutions were made up with double distilled water unless otherwise stated.

Filtered sea water	collected in Swansea Bay, allowed to settle for a few days, filtered through a GF/C filter, then a Millipore filter (0.45 µm pore size)

b. Aquil medium:

Although Erdshreiber medium has been used successfully for maintaining phytoplankton growth throughout this study, its definite composition has always been unknown. For the physiological experiments, a defined medium with known amounts of nutrients and which is suitable for the growth of the available phytoplankton was required. The basic idea for the preparation of such medium was to enrich synthetic sea water of known composition with additive nutrients. In addition to this type of medium synthetic media were tested. The media tested were: F_2 medium (Guillard and Ryther, 1962); ASP-2 (Provasoli et al., 1957); ARA medium (Kain and Fogg, 1958); Aquil (Morel et al., 1979); and the medium recommended by American Public Health Association (1980).

The suitability of these media for the growth of phytoplankton was tested. The growth was not satisfactory except in Aquil medium after few changes in its basic formula (Table 7.1).

ii. Phytoplankton species:

Most of the phytoplankton used in these laboratory experiments were obtained from the Plymouth laboratories via J. Sexton. Those species were Ditylum brightwellii, Thalassiosira sp., and Prorocentrum micans. Asterionella japonica was isolated from Swansea Bay during the course

Table 7.1 Composition of Aquil medium

SUBSTANCE	ORIGINAL FORMULA	PRESENT FORMULA
NaCl	420 mM	420 mM
$CaCl_2.2H_2O$	10.5 mM	10.5 mM
KBr	0.84 mM	0.84 mM
NaF	0.0714 mM	0.0714 mM
KCl	9.39 mM	9.39 mM
H_3BO_3	0.485 mM	0.485 mM
Na_2SO_4	28.87 mM	28.87 mM
$NaHCO_3$	2.38 mM	2.38 mM
$SrCl_2.6H_2O$	0.0638 mM	0.0638 mM
$MgCl_2.6H_2O$	54.6 mM	54.6 mM
$NaH_2PO_4.2H_2O$*	10 µM	25 µM
$NaNO_3$ *	100 µM	100 µM
Na_2SiF_6 *	12.5 µM	20 µM
$CuCl_2.2H_2O$	9.97×10^{-4} µM	4×10^{-2} µM
$Na_2MoO_4.2H_2O$	1.5×10^{-3} µM	3×10^{-2} µM
$CoCl_2.6H_2O$	2.5×10^{-3} µM	5×10^{-2} µM
$MnCl_2.4H_2O$	2.3×10^{-2} µM	0.9 µM
$ZnCl_2$	4×10^{-3} µM	8×10^{-2} µM
$FeCl_3$	4.51×10^{-1} µM	11.7 µM
Na_2.EDTA	5.0 µM	11.7 µM
Vitamin B-1	100 µg/l	100 µg/l
Vitamin B-12	0.55 µg/l	0.55 µg/l
Biotin	0.5 µg/l	0.5 µg/l

* Except when variable concentrations of nitrate, phosphate and silicate were needed

of this study.

Phytoplankton isolation

The method used here was based on the work of Knight-Jones (1951). It depends mainly on the dilution of sea water by Erdschreiber medium.

Procedure:

A preliminary dilution of 1/100 was made by adding 1 ml of the collected seawater to 99 ml of sterile sea water (previously autoclaved under 15 lb pressure/sq. inch for 20 minutes in a clean stoppered bottle and shaken gently several times).

Twenty test tubes each containing 9 ml Erdschreiber, plugged, and sterilized were arranged in four groups of five.

1 ml of the preliminary dilution was added to each test tube of the first group. After mixing the contents of each test tube well, 1 ml from each of these tubes was transferred to the corresponding tube in the second group. This process was repeated for the second, third and fourth group until a series of dilutions was achieved.

These tubes were then kept in a constant temperature room (20°C) under continuous illumination for a few days. The tubes were checked at different time intervals, and the one with unialgal growth was sub-cultured using Erdschreiber medium in conical flasks.

iii. Phytoplankton growth in culture media:

a. Cell counting:

Phytoplankton cells were counted either by Sedgewick rafter chamber or haemocytometer depending on cell size. The haemocytometer has been found to be much more suitable for counting the small phytoplankton

cells than the Sedgewick chamber.

b. Growth rate of phytoplankton species in Aquil and Erdschreiber media:

Replicate cultures of Prorocentrum micans, Ditylum brightwellii and Thalassiosira sp. were grown in Aquila and Erdschreiber media. P. micans and D. brightwellii cells were counted using Sedgewick rafter chamber, while Thalassiosira sp. was counted using the haemocytometer. The cell number of the three species was determined daily. The growth rate was determined as follows:

$$K = (\log \frac{N_2}{N_1}) \quad (\frac{3.322}{t})$$

where N_1 and N_2 are the cell concentrations at the beginning and end of a time period t
t = time in days

II. ^{14}C-fixation by phytoplankton in artificial media

A. Experiments conducted using full Aquil medium:

i. ^{14}C-fixation by phytoplankton species in unialgal cultures:

Three different species, Prorocentrum micans, Ditylum brightwellii and Thalassiosira sp. were used in these experiments. To a triplicate set of 125 ml bottles each containing 50 ml Aquil medium approximately the same number of P. micans cells was introduced. This process was repeated with the other species. They were then kept in a constant temperature room for four days. At the start of the fifth day, the cell number in each bottle was determined. At 3 minute intervals, 2 µci $NaH^{14}CO_3$ was added to each bottle and the counts were taken at zero time, and after four hours of incubation in the usual way (Chapter II). The ^{14}C-fixation was determined per 10^6 cells and per

surface area expressed as $\mu m^2.10^6$ cells hr^{-4}.

ii. ^{14}C-fixation by phytoplankton species in mixed algal culture: From the stock cultures of P. micans, D. brightwellii, and Thalassiosira sp., 1.425, 4.5 and 0.15 ml respectively were added to 100 ml full Aquil medium in a 500 ml conical flask to give approximately the same number of cells of each species in every flask. Five flasks were used, I, II, III, IV and V. Flasks numbers IV and V were used to determine the change in cell number during the course of the experiment. At 3 minute intervals, 10 µci $NaH^{14}CO_3$ was added to each of the flasks I, II and III. At zero time, 1 ml portions from each flask were filtered through 0.45, 20, 50 and 80 µm filters and the dpm was determined for each filter.

The five cultures were kept in a constant temperature room under growing conditions for 7 days. Every 48 hours the cell number was counted and the above filtration procedures were repeated.

B. Experiments conducted using media with variable nitrate concentration:

i. Effect of nitrate on the growth of and ^{14}C-fixation by Thalassiosira sp.:

Thalassiosira sp. grown in Erdschreiber medium was centrifuged at 5000 rpm, and washed twice with N-free Aquil medium. These washed Thalassiosira cells were resuspended in N-free Aquil medium to give a concentration of 192,500 cells ml^{-1}. This suspension was used to inoculate 20 x 125 ml bottles each containing 100 ml Aquil medium. These bottles were separated into two groups each with ten bottles. In each group duplicates were made of the following nitrate concentrations:

Control; 1 µg at Nl^{-1}; 10 µg at Nl^{-1}; 50 µg at Nl^{-1}; and 100 µg at Nl^{-1}. To each of the 20 bottles, inoculum from the cell suspension was added to give a final cell count of ca. 3-5 x 10^3 cells ml^{-1}. The bottles of the first group were used for the growth experiment and the bottles of the second group were used for the ^{14}C-fixation experiments.

On the first day of the experiment the number of cells was counted in the first group, and to the second group 5 µci $NaH^{14}CO_3$ was added to each culture. As soon as the radioactive carbon was added, 10 ml were filtered from each bottle and the ^{14}C-fixation was measured at zero time.

These cultures were kept in the constant temperature room at 20°C and continuously illuminated by fluorescent light. Every 24 hours the number of cells and ^{14}C-fixation were determined.

ii. Effect of nitrate on ^{14}C-fixation by N-starved *Thalassiosira sp.*; *Asterionella japonica*:

In this type of experiment the algal cells were nitrogen starved by washing in N-free medium and then grown at constant temperature for 48 hours. Two media were used, Aquil medium and Erdschreiber medium. N-free Erdschreiber medium was obtained by growing *Phaeodactylum tricornutum* cells in Erdschreiber medium to exhaust the ambient nitrogen in the sea water. After complete exhaustion of nitrogen, the Erdschreiber medium was filtered through GF/C filters and the filtrate was enriched with phosphate and silicate. In all the experiments, duplicates were made for each culture.

a. *Thalassiosira sp.*:

Starved cells were inoculated into a series of flasks containing 100 ml

N-free Aquil medium and varying nitrate concentrations (0, 0.1, 1, 10, 100 µg at l^{-1}) to give final concentration of approximately 3 x 10^3 cells ml^{-1}. To each of these flasks 5 µci $NaH^{14}CO_3$ were added. The ^{14}C-fixation was measured at zero time and after four hours of incubation.

b. Asterionella japonica:

Starved cells were inoculated into a series of flasks containing 100 ml N-free Erdschreiber medium and varying nitrate concentrations (0, 5, 20, 50, 100 µg at Nl^{-1}) to give a final concentration of approximately 200 cells ml^{-1}. To each of these flasks 4 µci $NaH^{14}CO_3$ were added. The ^{14}C-fixation was measured at zero time and after four hours of incubation.

The following experiments were developed, designed and based on the previous experiments (ii.a and ii.b). In the new experiments, Thalassiosira sp. was preferred to A. japonica because it has been found much easier to grow, maintain and count.

iii. Effect of nitrate on ^{14}C-fixation by N-starved Thalassiosira sp. at different time intervals:

Starved cells of Thalassiosira sp. were inoculated into a series of bottles containing 100 ml N-free Aquil medium and varying nitrate concentrations (0, 1, 10, 100, 1000 µg at l^{-1}) to give final concentrations of approximately 10 x 10^3 cells ml^{-1}. To each of these bottles 5 µci $NaH^{14}CO_3$ were added. The ^{14}C-fixation was measured every hour for four hours.

iv. ^{14}C-fixation by Thalassiosira sp. cells previously adapted to different nitrate concentrations:

An experiment similar to the one described above (iii) was carried out. In this experiment, the cells were adapted to their new nitrate concentration for 24 hours before the addition of $NaH^{14}CO_3$. The ^{14}C-fixation was measured after four hours of incubation only.

v. Effect of nitrate on chlorophyll a content of N- starved Thalassiosira sp.:

Thalassiosira sp. cells were centrifuged and washed in N-free Aquil medium. Approximately the same number of washed cells were inoculated to bottles containing 100 ml Aquil medium with the following range of nitrate concentrations: 0, 5 and 500 µg at Nl^{-1}. The cell number was determined for each culture prior to every filtration. At zero time, 5 ml of each culture was filtered through GF/C filter for chlorophyll a determination. The cultures were incubated in a constant temperature room at 20°C and the filtration process was repeated at 7, 24, 48 and 72 hours. Chlorophyll a content was determined following the method of Strickland and Parsons (1972) and it was expressed as mg chlorophyll a per 10^6 cells.

III. Sea water enrichment and the ^{14}C-fixation by phytoplankton size fractions

In these experiments, sea water samples from Station A were enriched with growth nutrient. The aim was to investigate the effect of nutrient enrichment on ^{14}C-fixation by phytoplankton size fractions. Three experiments were carried out, each with a different combination of nutrient concentrations. The nutrients added and their concentrations in each experiment are mentioned in Table 7.2.

Table 7.2

CULTURE	EXPERIMENT 1*	EXPERIMENT 2**	EXPERIMENT 3***
Control	No addition	No addition	No addition
Nitrate	+5 µg at l^{-1}	+50 µg at l^{-1}	+50 µg at l^{-1}
	+50 µg at l^{-1}	+500 µg at l^{-1}	+500 µg at l^{-1}
Phosphate	+0.5 µg at l^{-1}		
	+10 µg at l^{-1}		

* The sea water used in this experiment contained 13.65 µg at Nl^{-1} and 0.835 µg at Pl^{-1}.

** The sea water used in this experiment contained 31.4 µg at Nl^{-1} and 1.2 µg at Pl^{-1}. Phosphate was added to all cultures to give final concentration of 20 µg at Pl^{-1}.

*** To all cultures phosphate was added to give final concentration of 20 µg at Pl^{-1}, and silicate to give final concentration of 50 µg at Sil^{-1}.

In these experiments 500 ml of the collected sea water was poured into a 2 litre conical flask. To this sample the required amount of nutrient(s) was added. Triplicates were made for each culture and the flasks were then kept continuously illuminated in a constant temperature room at 20°C. On the day of filtration, 50 ml from each culture was poured into a 125 ml bottle. At zero time, 2 µci $NaH^{14}CO_3$ was added to this sample. After 4 hours of incubation in the usual way (Chapter II), 3 x 10 ml portions from each sample were filtered through 0.45, 5 and 20 µm filters. The radioactivity was then counted (Chapter II). In experiment no.3, the nitrate uptake by phytoplankton was measured through its loss from the medium. On the day of ^{14}C-fixation measurement, a 5 ml portion from each culture was filtered through GF/C filters using a syringe filter holder and the filtrate was stored

frozen in 5 ml capped plastic containers. Later, they were thawed and the nitrate concentrations were determined using the method described by Cawse (1967). This method is described below:

a. Reagents:

Perchloric acid, 5% v/v

Sulphamic acid, 2% w/v

b. Procedure:

Take 1 ml of the filtered sample in a test tube, add 1 ml of sulphamic acid solution and allow the mixture to stand for 2 minutes. Dilute to 10 ml with 5% v/v perchloric acid, and measure the absorbance at 210 nm with 1 cm silica cells. Correct for blank.

c. Calibration curve:

Nitrate standard solution was prepared (0.1 µg at N ml^{-1}). This was diluted to 0.75, 0.5 and 0.25 µg at N ml^{-1}. Triplicates of these dilutions were prepared, and to each ml of the standard 1 ml sulphamic acid was added followed by 8 ml perchloric acid after 2 minutes. The optical densities obtained were very close to those mentioned in the original work (Cawse, 1967). From the present results an optical factor was calculated (F = 1390). See Figure 7.1, Table 7.3.

Figure 7.1 Nitrate calibration curve

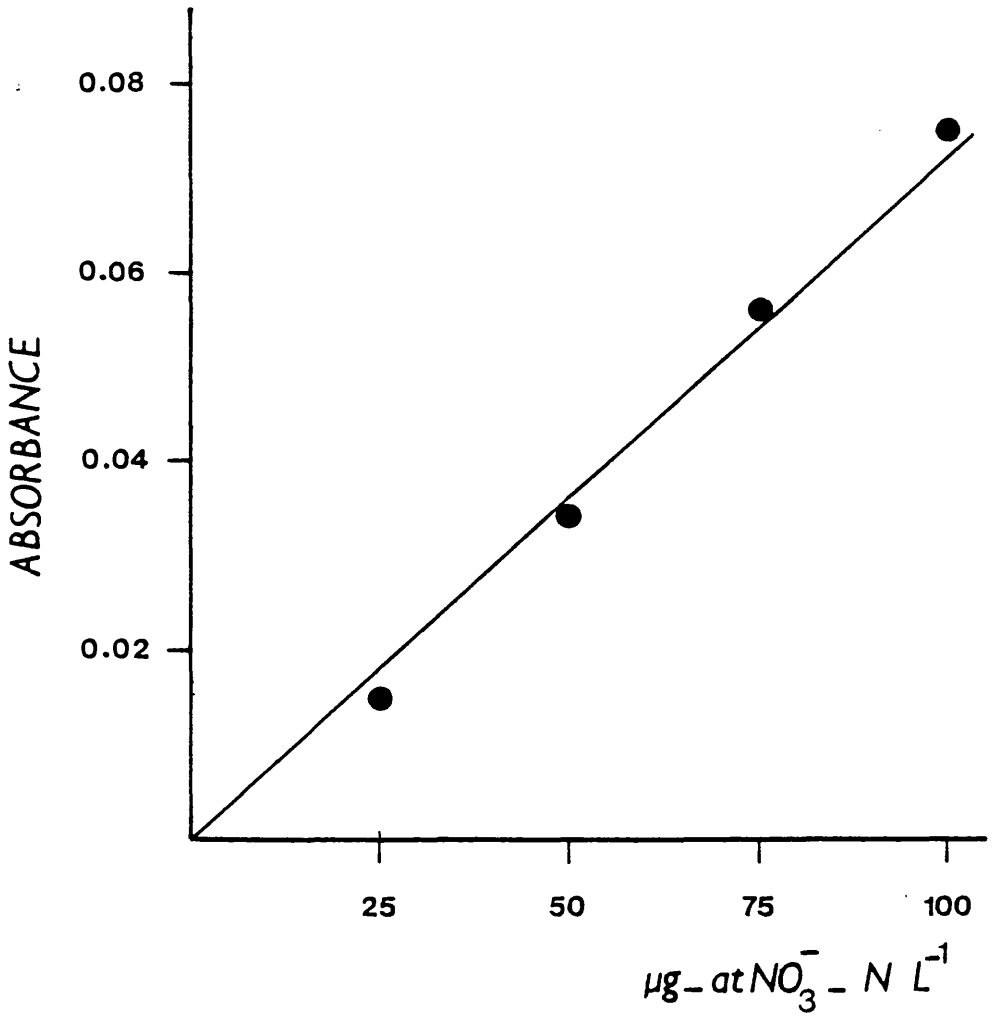

0.08
0.06
0.04
0.02
ABSORBANCE
25
50
75
100
μg-at NO3- N L-1

Table 7.3 Results of nitrate calibration curve

CONCENTRATION	AVERAGE Es - Eb	CALIBRATION FACTOR
25 µg at 1^{-1}	0.0155	1613
50 µg at 1^{-1}	0.0345	1449
75 µg at 1^{-1}	0.056	1339
100 µg at 1^{-1}	0.075	1335

Es : Extinction of sample

Eb : Extinction of blank

RESULTS

I. Culture Media and Phytoplankton Growth

Growth rate of phytoplankton species in Aquil and Erdscreiber medium: The growth rates of Thalassiosira sp., Prorocentrum micans and Ditylum brightwellii each in Erdshreiber and Aquil media are shown in Figures 7.2, 7.3 and 7.4. The highest growth rate of Thalassiosira in Erdschreiber was found within the first 24 hours of growth (k = 1.7). This was followed by a gradual decline until it reached -0.02 doubling/ day in the period between days 5 and 6. In Aquil medium the growth of Thalassiosira reached its peak after the first day of incubation (k = 1.72). This peak was followed by a rapid decrease in the value of growth rate until it reached it lowest value (k = 0.02) between the fifth and sixth days.

The growth of P. micans in Erdshcreiber (Fig. 7.3) exhibits a pattern with a high growth rate value in the first time interval (k = 1.0) followed by a drop down to 0.1 in the second time interval.

Figure 7.2 Growth rate of Thalassiosira sp.

in a: Erdschreiber medium
b: Aquil medium

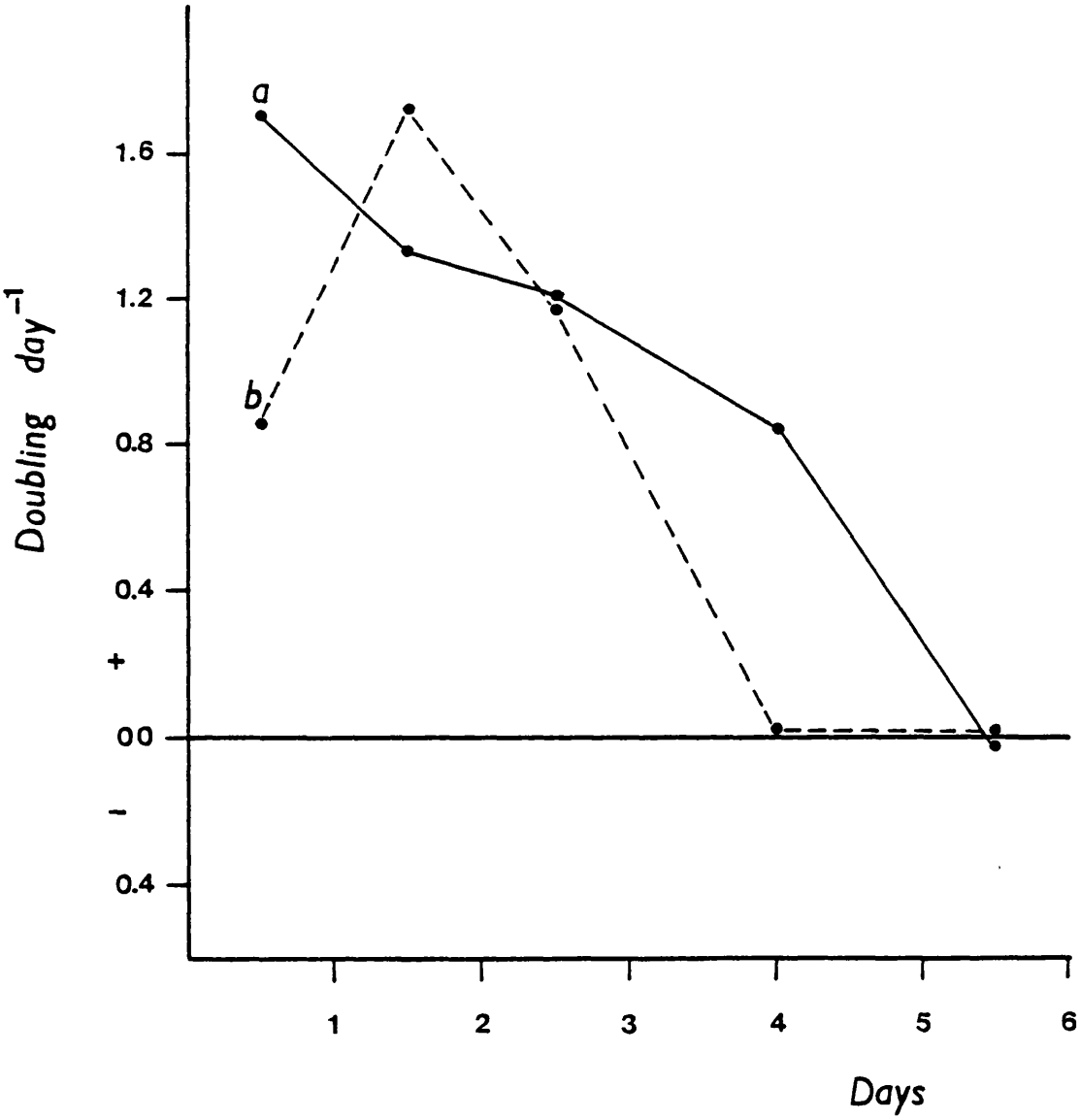

a
b
1.6
1.2
0.8
0.4
+
00
−
0.4
Doubling day⁻¹
1
2
3
4
5
6
Days

Figure 7.3 Growth rate of Prorocentrum micans

in a: Erdschreiber medium
b: Aquil medium

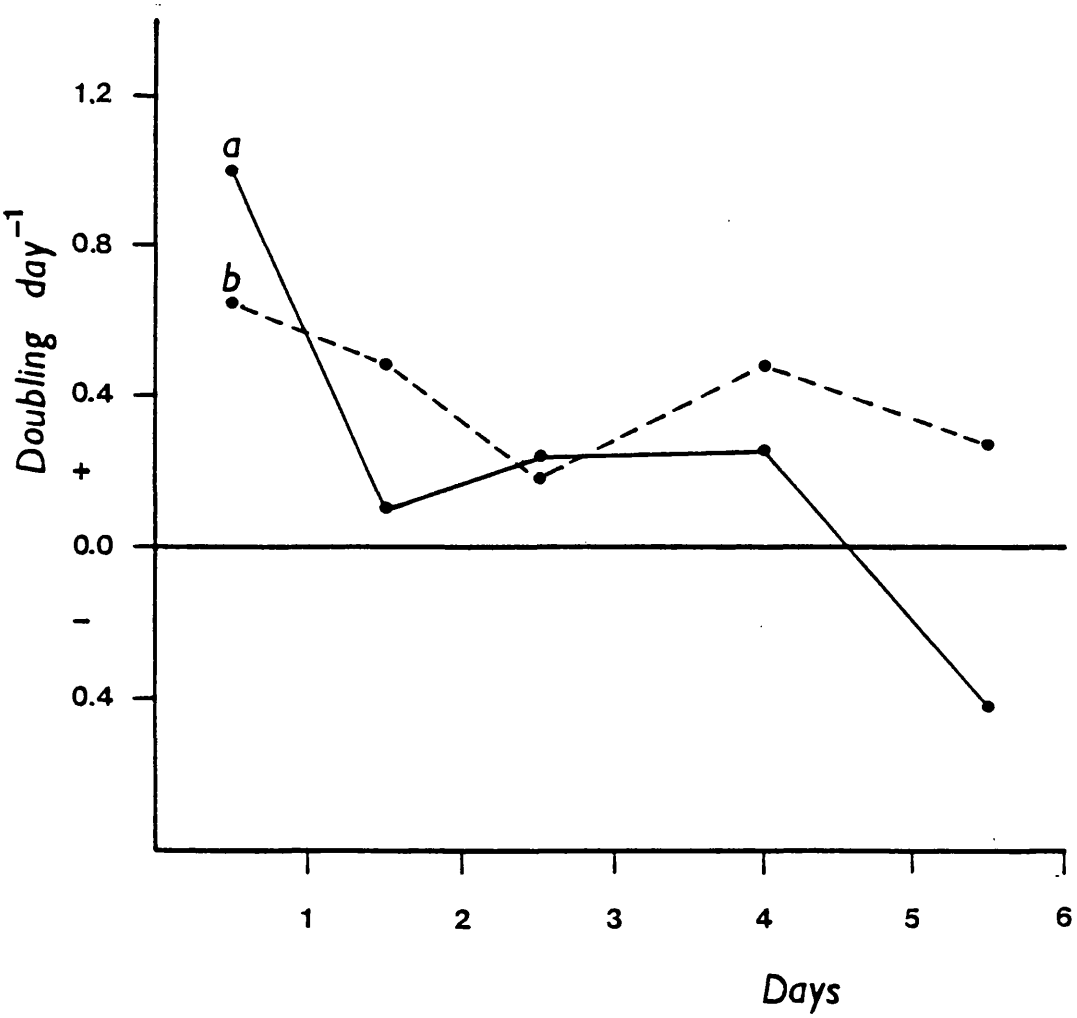
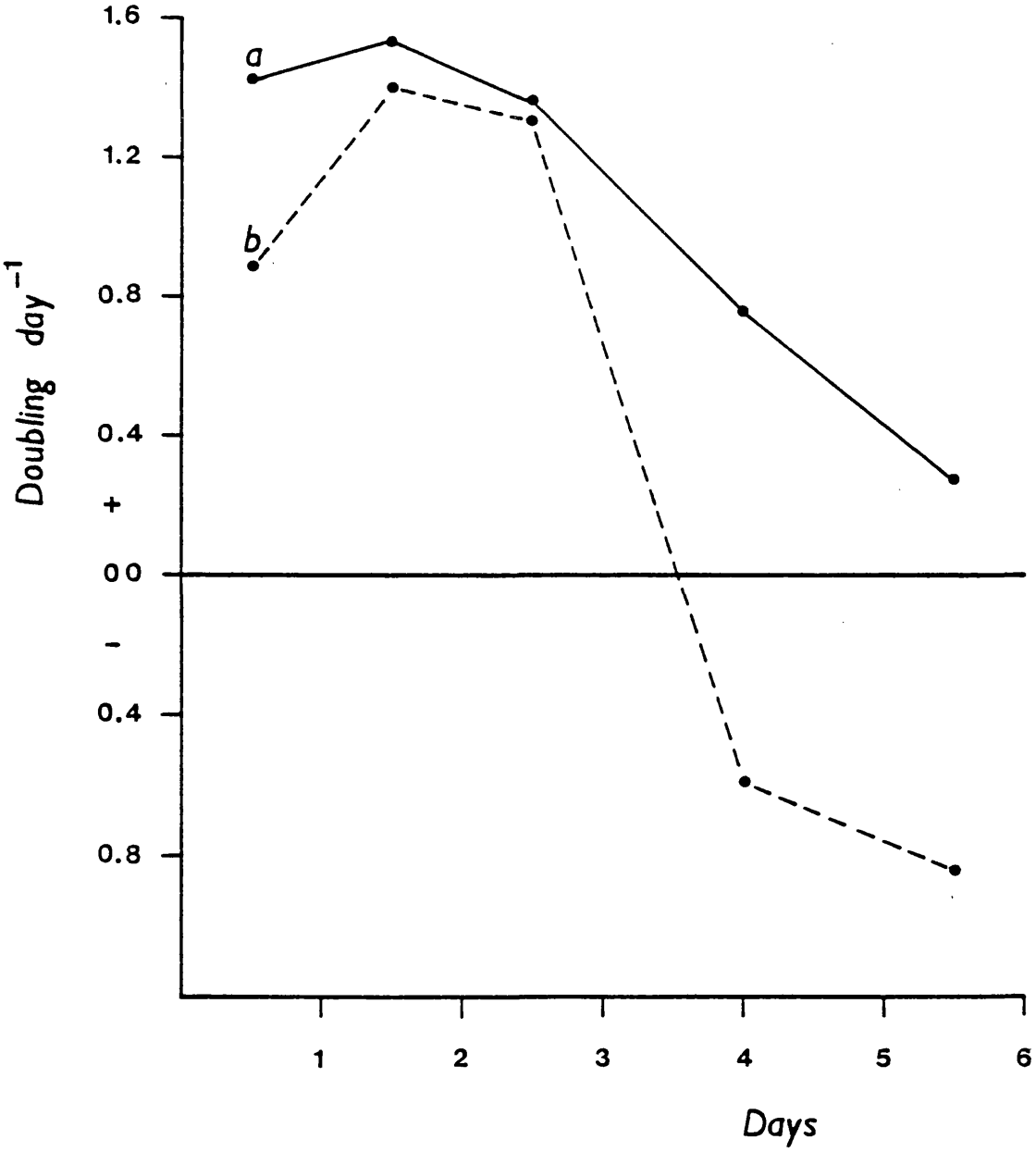

1.2
0.8
0.4
+
0.0
−
0.4
Doubling day^{-1}
a
b
1
2
3
4
5
6
Days

Figure 7.4 Growth rate of Ditylum brightwellii

in a: Erdschreiber medium
b: Aquil medium


1.6
1.2
0.8
0.4
+
00
-
0.4
0.8
a
b
Doubling day^{-1}
1
2
3
4
5
6
Days

Between the second and the fifth day, the growth rate was ca. 0.22. This value dropped to -0.42 in the period between the fifth and sixth day. To a certain extent the growth rate of P. micans in Aquil medium was similar to that in Erdschreiber. The highest growth rate value of 0.64 in the first time interval decreased gradually to 0.18 in the third time interval. After an increase to 0.48 in the period between the third and fifth day the growth rate decreased to 0.27 in the final period. The growth rate of Ditylum brightwellii in Erdschreiber and Aquil media is illustrated in Figure 7.4. The growth rate in Erdschreiber medium reached its highest value in the second time interval (k = 1.42). There was a gradual decrease down to 0.27 in the last time interval between the fifth and sixth day. The growth rate pattern of D. brightwellii in Aquil medium was similar to that in Erdschreiber medium. The highest value (k = 1.4) which was found in the second time interval was followed by a sharp decline after a short period down to -0.6 in the period between the third and fifth day. This was followed by a further decrease to -0.84 in the last time interval.

II. ^{14}C-fixation by Phytoplankton in Artificial Media

A. Experiments conducted using full Aquil medium:

i. ^{14}C-fixation by phytoplankton species in unialgal cultures:

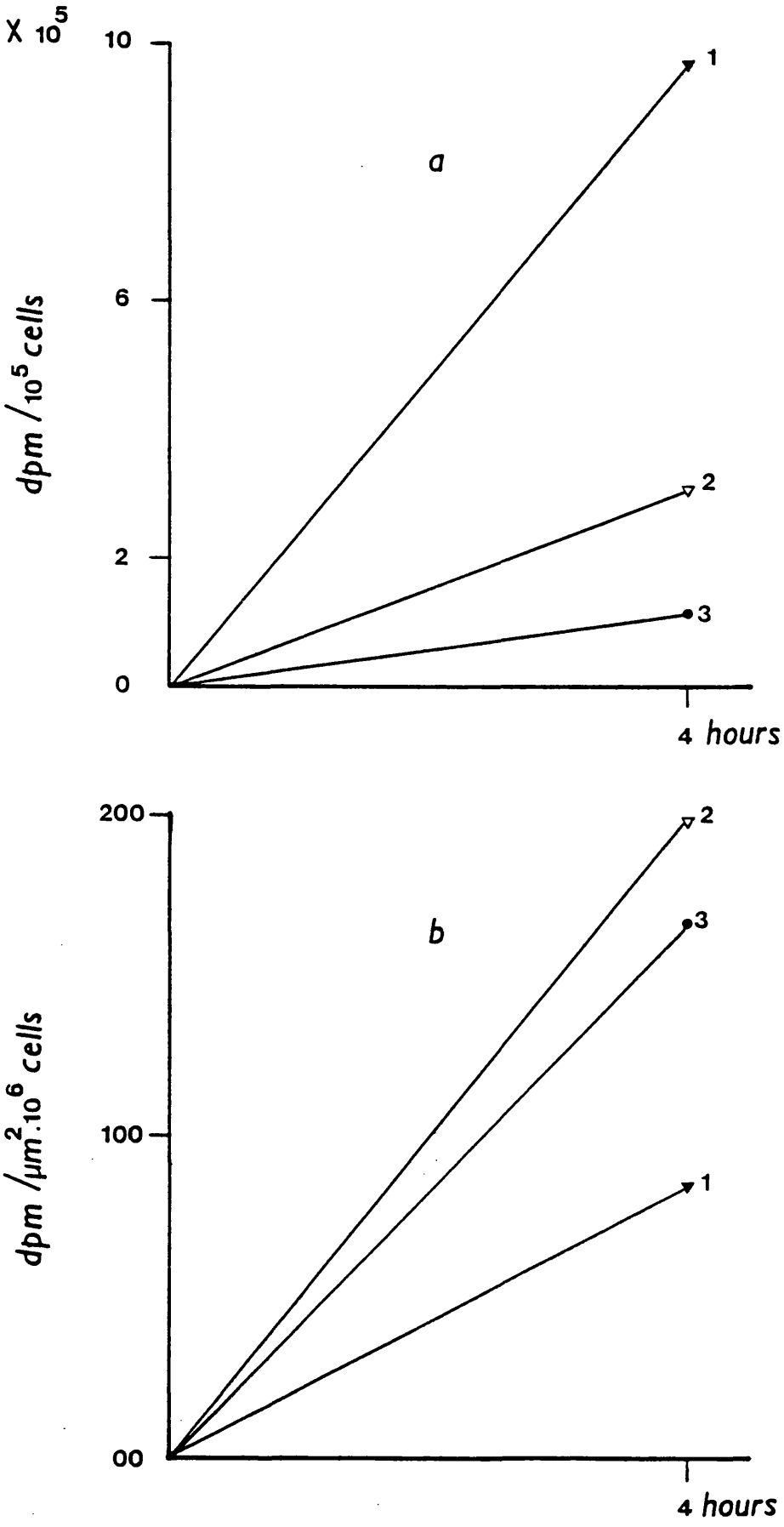
Figure 7.5a illustrates the fixation of ^{14}C- by three different phytoplankters, Thalassiosira sp., Prorocentrum micans, and Ditylum brightwellii. The fixation is expressed as dpm/10^6 cells. D. brightwellii had the highest fixation value of 9.65×10^5 dpm/10^6 cells. P. micans followed with a value of 3×10^5 dpm/10^6 cells and Thalassiosira sp. with the lowest value of the three of 1.15×10^5

Figure 7.5a ^{14}C-fixation by phytoplankton in unialgal cultures (dpm/10^6 cells)

1. D. brightwellii
2. P. micans
3. Thalassiosira sp.

Figure 7.5b ^{14}C-fixation by phytoplankton in unialgal cultures (dpm/$\mu m^2.10^6$ cells)

1. D. brightwellii
2. P. micans
3. Thalassiosira sp.


X 10^5
10
6
2
0
dpm /10^5 cells
a
1
2
3
4 hours
200
100
00
dpm /μm^2.10^6 cells
b
2
3
1
4 hours

dpm/10^6 cells. Figure 7.5b illustrates the same results when expressed as dpm/$\mu m^2.10^6$ cells. In this case the order of fixation has changed with P. micans at the top with ca. 200 dpm/$\mu m^2.10^6$ cells. It was followed by Thalassiosira sp. with 166 dpm/$\mu m^2.10^6$ cells. D. brightwellii followed with a value of 84 dpm/$\mu m^2.10^6$ cells.

ii. ^{14}C-fixation by phytoplankton species in mixed algal culture:
In this part, three phytoplankters viz. Thalassiosira sp., P. micans and D. brightwellii were grown in Aquil medium. The growth of these species in mixed culture is illustrated in Figure 7.6. From an initial cell concentration of ca. 750 cell ml^{-1} Thalassiosira increased during the 7-days-long experiment reaching a maximum of 37,500 cell ml^{-1} on the fifth day. Although P. micans and D. brightwellii started with about the same initial cell concentration, the growth rates of both species were much slower than that of Thalassiosira. P. micans cell concentration remained at the same level from the second day of the experiment until the end which was ca. 1500 cell ml^{-1}. D. brightwellii growth curve followed a similar pattern to that of P. micans but in this case at a slightly higher level of ca. 3750 cell ml^{-1}.

Figure 7.7 illustrates the increase in cell surface area of the three species. D. brightwellii total surface area increased very rapidly from ca. 8 x 10^6 μm^2 up to 50.5 x 10^6 μm^2 on the fifth day. Thalassiosira sp. total surface area increased from 0.5 x 10^6 μm^2 up to 26.5 x 10^6 μm^2 on the fifth day. The total cell surface area of P. micans increased at a very slow rate. From 0.85 x 10^6 μm^2 of the initial P. micans cell concentration, the total surface area remained around 1.9 x 10^6 μm^2 reaching its highest value of ca. 2.5 x 10^6 μm^2 on the fifth day.

Figure 7.6 Growth of phytoplankton species in mixed culture

1. D. brightwellii
2. P. micans
3. Thalassiosira sp.

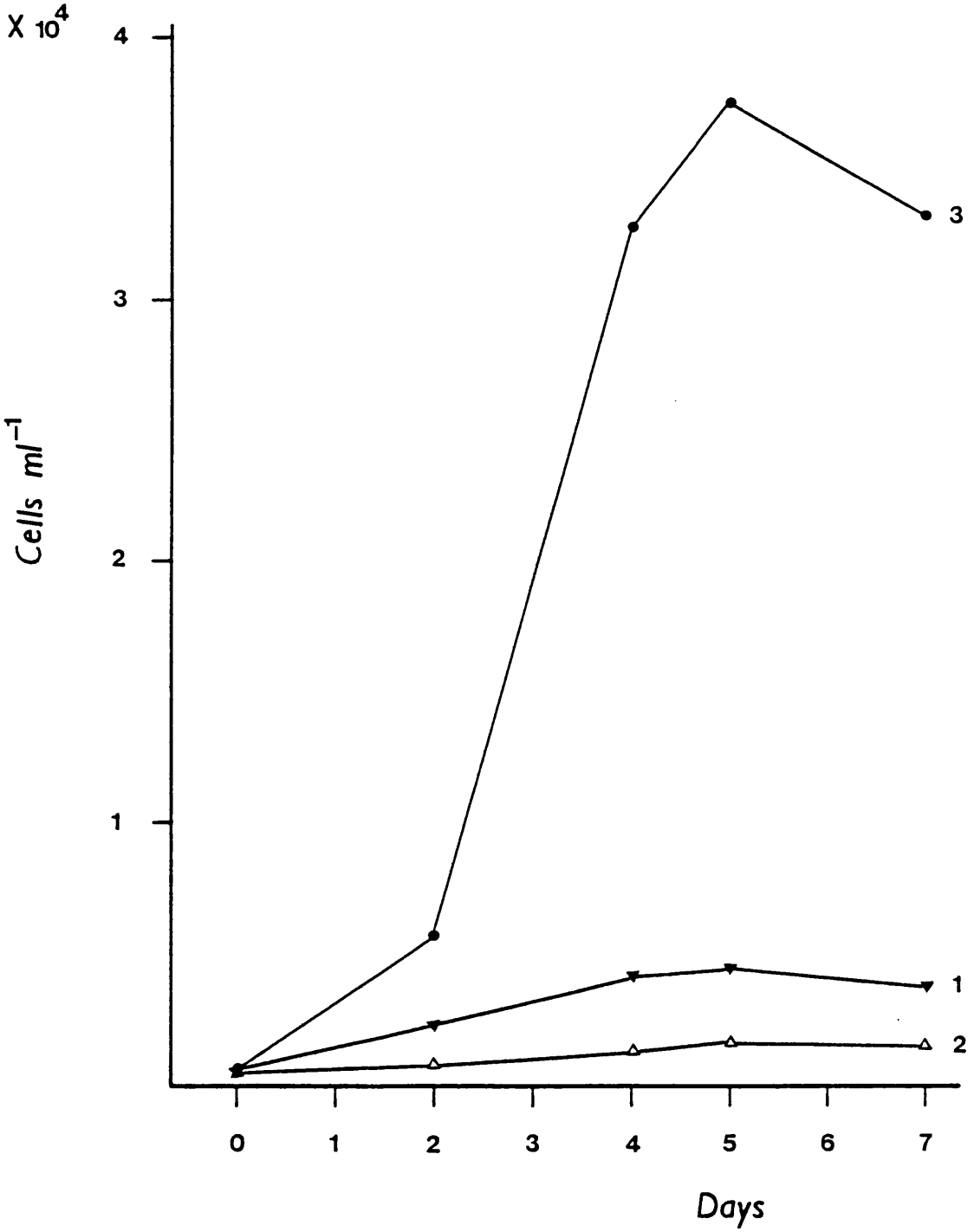

X 10^4
4
3
2
1
Cells ml^-1
3
1
2
0
1
2
3
4
5
6
7
Days

Figure 7.7 Change of phytoplankton surface area in mixed culture

1. D. brightwellii
2. P. micans
3. Thalassiosira sp.

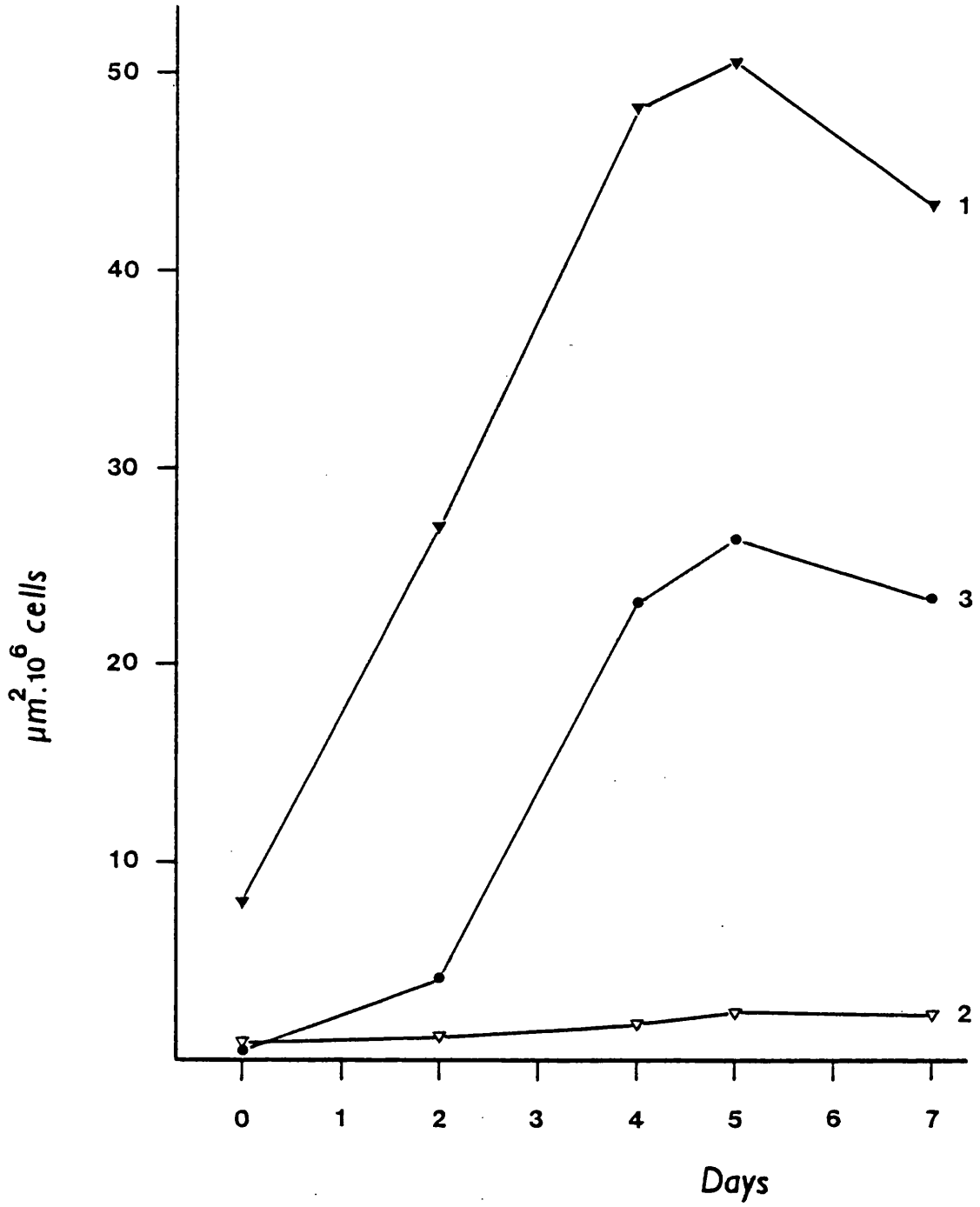

50
40
30
20
10
$\mu m^2.10^6$ cells
1
3
2
0
1
2
3
4
5
6
7
Days

Figure 7.8 illustrates the fixation of $NaH^{14}CO_3$ by the different size fractions of the mixed culture. The initial fixation at zero time by each species was < 100 dpm. The fixation by the size class 0.45-20 µm was lowest on the second day. This increased steadily and reached its highest fixation on the fifth day. At that point the fixation by this size class was representing ca. 35% of the total ^{14}C-fixation. On the second day the size class of organisms > 50 µm showed the second highest fixation, but thereafter their role was taken over by the organisms in the size class 0.45-20 µm. The highest fixation by the phytoplankton in the size class > 50 µm was on the fourth day of the experiment where they represented 27% of the total ^{14}C-fixation. Throughout the experiments, members of the size class 20-50 µm dominated the population reaching their highest fixation on the fifth day with 40% of the total. ^{14}C-fixation by each species was calculated by working out a correction for the differential retention by the filters i.e. calculating the percentage of each species retained by the different filters. This was achieved by filtering 1 ml from selected mixed culture used for determining cell concentration. The results obtained show that 90% of Thalassiosira sp. pass through the 20 µm filter, 25% of D. brightwellii pass through the 50 µm filter. When the results of the ^{14}C-fixation by the different size classes shown in Figure 7.8 were corrected to give the fixation by each species, Figure 7.9 was achieved. This figure shows that Thalassiosira and D. brightwellii represented the major contributors to the ^{14}C-fixation throughout the experiment. But when the fixation was expressed as dpm/10^6 cells of each species (Fig. 7.10), P. micans contributed the major fixation throughout the experiment with D. brightwellii in the second place. When the fixation by the unit

Figure 7.8 ^{14}C-fixation by different size classes in mixed culture

1. 0.45 - 20 µm
2. 20 - 50 µm
3. > 50 µm

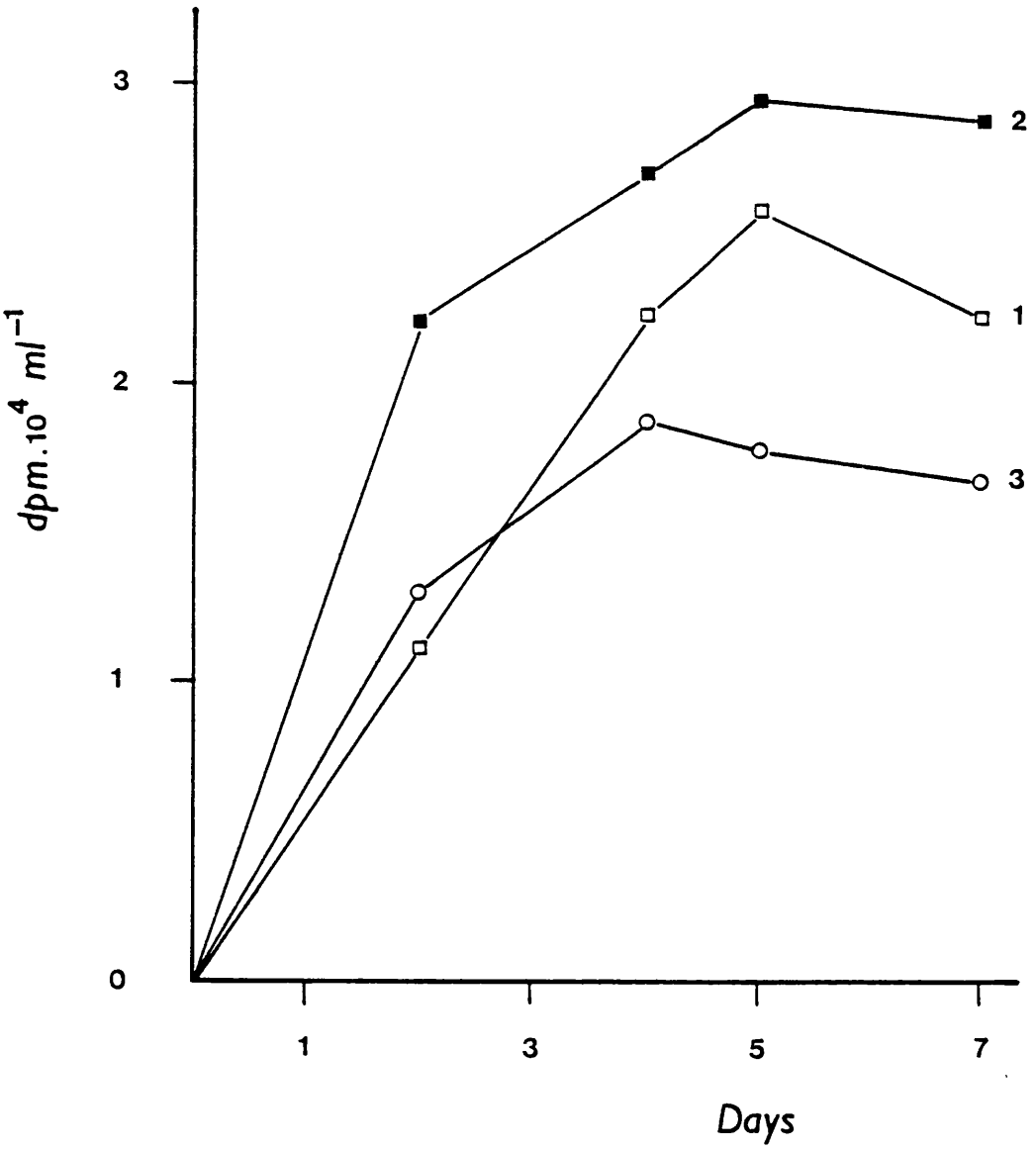

3
2
1
0
dpm.10^4 ml^-1
1
3
5
7
Days
2
1
3

Figure 7.9 ^{14}C-fixation by phytoplankton in mixed culture

1. D. brightwellii
2. P. micans
3. Thalassiosira sp.

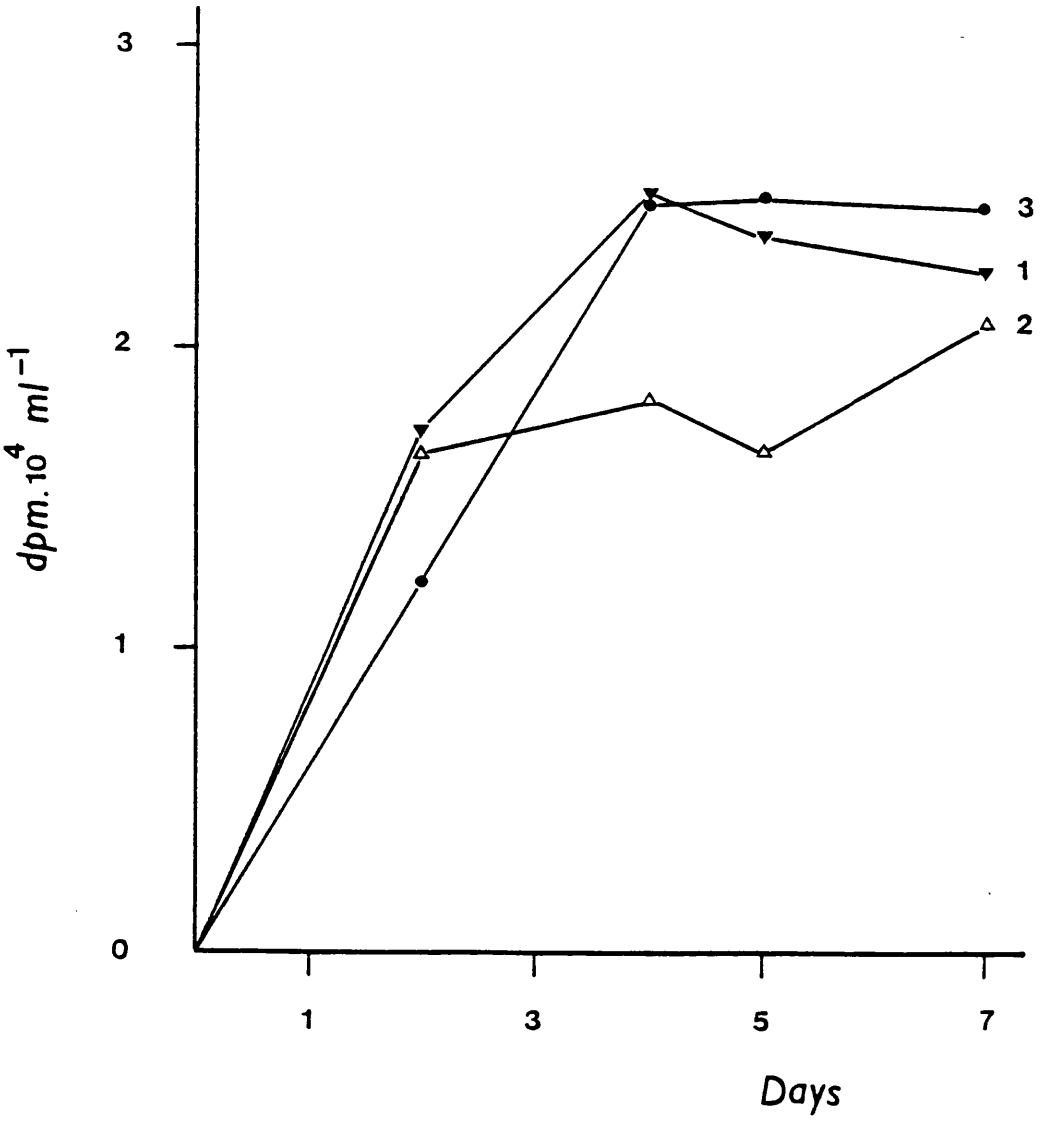
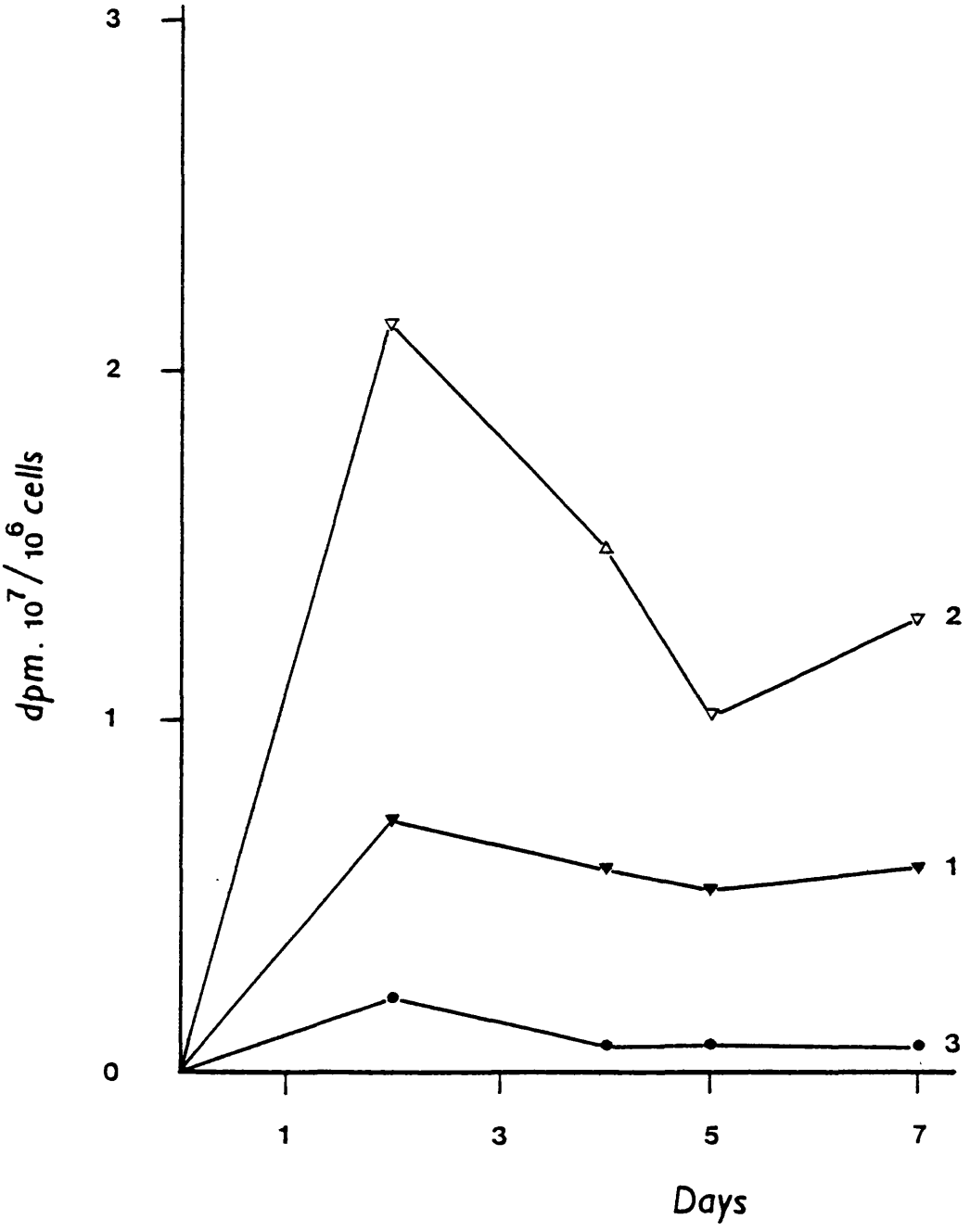

3
2
1
0
dpm.10^4 ml^-1
1
3
5
7
Days
3
1
2

Figure 7.10 ^{14}C-fixation by phytoplankton in mixed culture

1. D. brightwellii
2. P. micans
3. Thalassiosira sp.


3
2
1
0
dpm. 10^7 / 10^6 cells
2
1
3
1
3
5
7
Days

surface area was considered (Fig. 7.11), P. micans represented the highest fixation/$\mu m^2.10^6$ cells. It was followed by Thalassiosira sp.. Figure 7.11 shows that both P. micans and Thalassiosira sp. had their maximum fixation/unit surface area on the second day, which was during the exponential growth phase.

B. Experiments conducted using media with variable nitrate concentrations:

i. Effect of nitrate on the growth of, and ^{14}C-fixation by Thalassiosira sp.:

a. Growth (Fig. 7.12):

The cell number increased in the control on the second day from 0.85 x 10^4 cell ml^{-1} to 2.8 x 10^4 cell ml^{-1}. The cell number remained at this level to the end. At the 1 µg at l^{-1} concentration, the cell number increased from 1.4 x 10^4 cell ml^{-1} on the second day to 4.1 x 10^4 cell ml^{-1} on the fifth day. After reaching this value, the cell number decreased slightly at the end of the experiment. At the 10 µg at l^{-1} concentration, the cell number increased gradually from 0.5 x 10^4 cell ml^{-1} at the start of the experiment to 5.4 x 10^4 cell ml^{-1} on the fifth day. This was followed by a drop to 4.2 x 10^4 cell ml^{-1} on the last day of the experiment. At the 50 µg at l^{-1} concentration, the cell number increased from 0.33 x 10^4 cell ml^{-1} on the first day to 8.8 x 10^4 cell ml^{-1} on the fifth day. This was followed by a drop to 6.33 x 10^4 cell ml^{-1} on the sixth day. At the 100 µg at l^{-1} concentration, the cell number increased from 0.5 x 10^4 cell ml^{-1} on the first day to 12.55 x 10^4 cell ml^{-1} on the fifth day. This was followed by a drop to 8.8 x 10^4 cell ml^{-1} on the sixth day.

Figure 7.11 ^{14}C-fixation by phytoplankton in mixed culture ($dpm/\mu m^2.10^6$ cells)

1. D. brightwellii
2. P. micans
3. Thalassiosira sp.

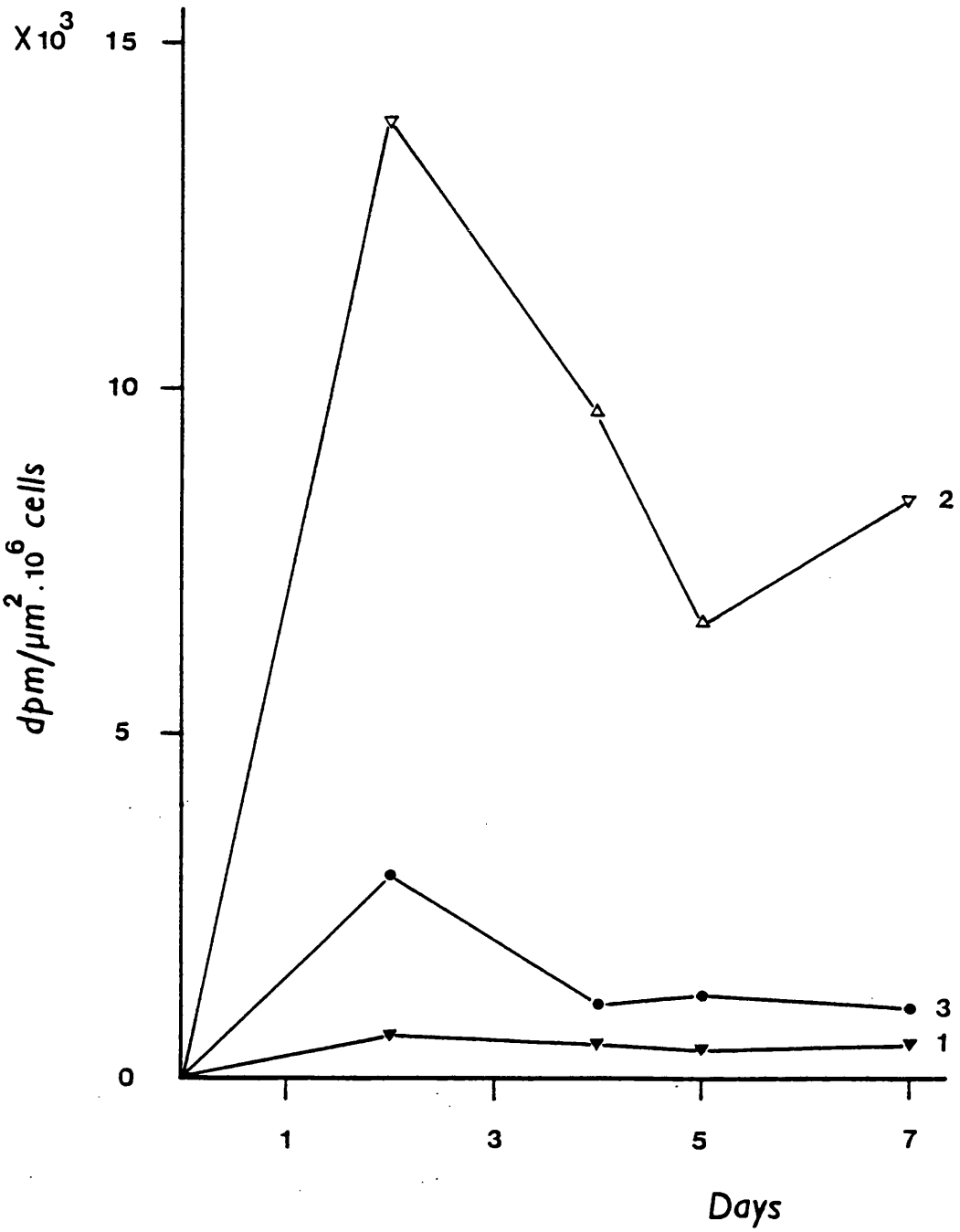
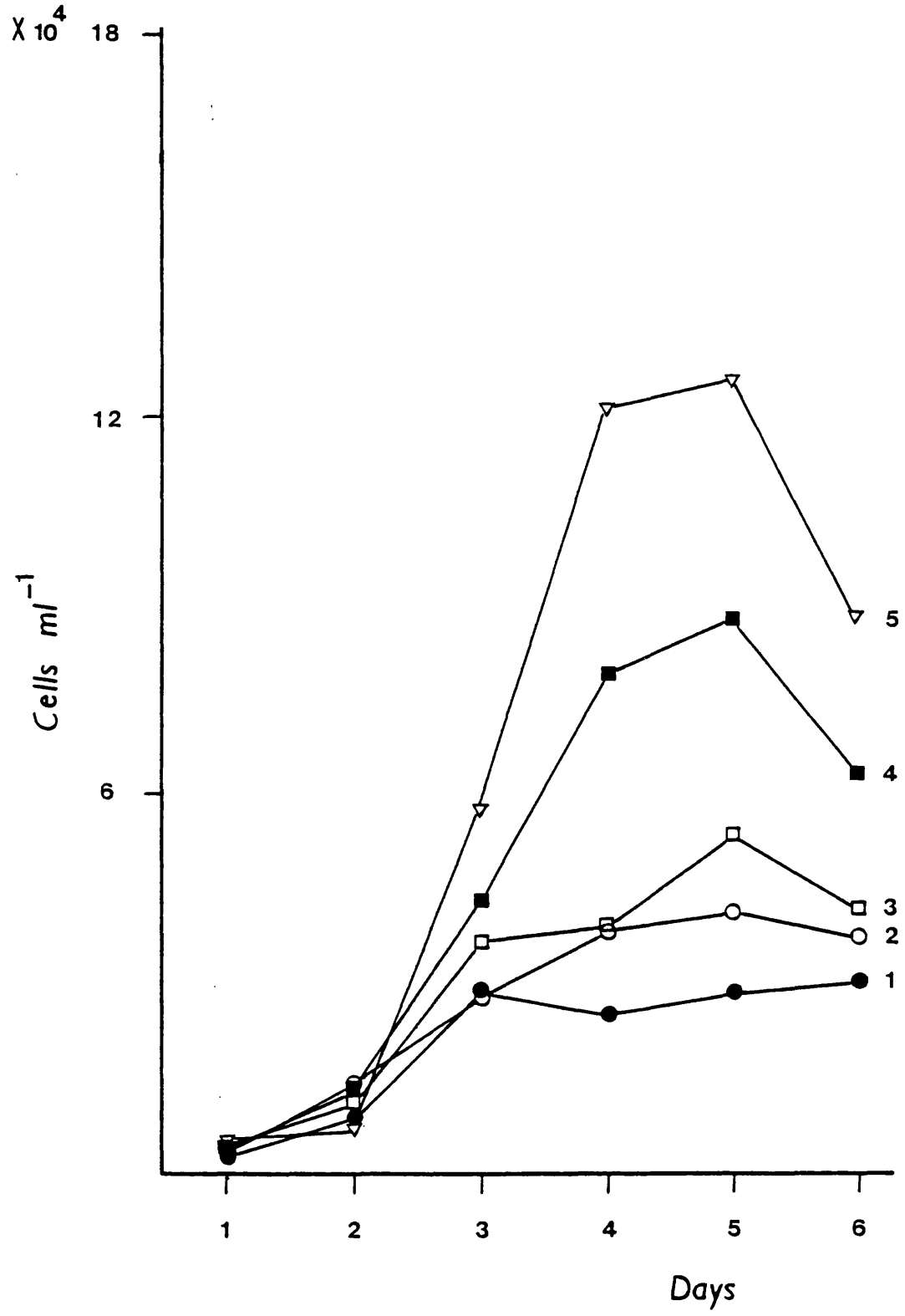

X 10^3
15
10
5
0
dpm/μm^2 . 10^6 cells
1
3
5
7
Days
2
3
1

Figure 7.12 Effect of nitrate on the growth of Thalassiosira sp.

1. control
2. 1 µg at Nl^{-1}
3. 10 µg at Nl^{-1}
4. 50 µg at Nl^{-1}
5. 100 µg at Nl^{-1}


X 10^4
18
12
6
Cells ml^-1
1
2
3
4
5
6
Days
5
4
3
2
1

b. ^{14}C-fixation (Fig. 7.13):

The ^{14}C-fixation by Thalassiosira cells grown in control, 1 µg at Nl^{-1}, and 10 µg at Nl^{-1} were similar and progressed at the same rate (Fig. 7.13). From an initial value of ca. 3.7×10^3 dpm ml^{-1} at the beginning of the experiment, the ^{14}C-fixation of the three concentrations increased up to 1.022×10^6 dpm ml^{-1} (control), 1.077×10^6 dpm ml^{-1} (1 µg at l^{-1}), and 1.118×10^6 dpm ml^{-1} (10 µg at l^{-1}) at the end of the experiment. The ^{14}C-fixation at the 50 µg at l^{-1} increased gradually from 3.3×10^3 dpm ml^{-1} at the beginning of the experiment to 1.538×10^6 dpm ml^{-1} at the end. The ^{14}C-fixation at the 100 µg at l^{-1} increased rapidly from 3.1×10^3 dpm ml^{-1} at the beginning to 2.601×10^6 dpm ml^{-1} at the end of the experiment.

ii. Effect of nitrate on ^{14}C-fixation by N-starved cells of Thalassiosira sp.; Asterionella japonica:

a. Thalassiosira sp. (Fig. 7.14):

At 0 µg at Nl^{-1} (control), ^{14}C-fixation value was 3.36×10^4 dpm/10^6 cells. At 0.1 µg at Nl^{-1}, ^{14}C-fixation value was 3.4×10^4 dpm/10^6 cells. ^{14}C-fixation at 1 µg at Nl^{-1} was 2.975×10^4 dpm/10^6 cells. At 10 µg at Nl^{-1}, ^{14}C-fixation was 2.025×10^4 dpm/10^6 cells, and at 100 µg at Nl^{-1} it was 1.15×10^4 dpm ml^{-1}.

b. Asterionella japonica (Fig.7.15):

At 0 µg at Nl^{-1} (control), the ^{14}C-fixation was at its highest value (3.02×10^3 dpm ml^{-1}). At 5 µg at l^{-1}, it was 1.74×10^3 dpm ml^{-1}. At 20 µg at l^{-1} the ^{14}C-fixation was 0.26×10^3 dpm ml^{-1}. At 50 µg at l^{-1} it was 0.18×10^3 dpm ml^{-1}, and the ^{14}C-fixation by nitrogen-starved A. japonica incubated at 100 µg at Nl^{-1} was 0.22×10^3 dpm ml^{-1}.

Figure 7.13 Effect of nitrate on the ^{14}C-fixation by *Thalassiosira sp.*

1. control
2. 1 µg at Nl^{-1}
3. 10 µg at Nl^{-1}
4. 50 µg at Nl^{-1}
5. 100 µg at Nl^{-1}

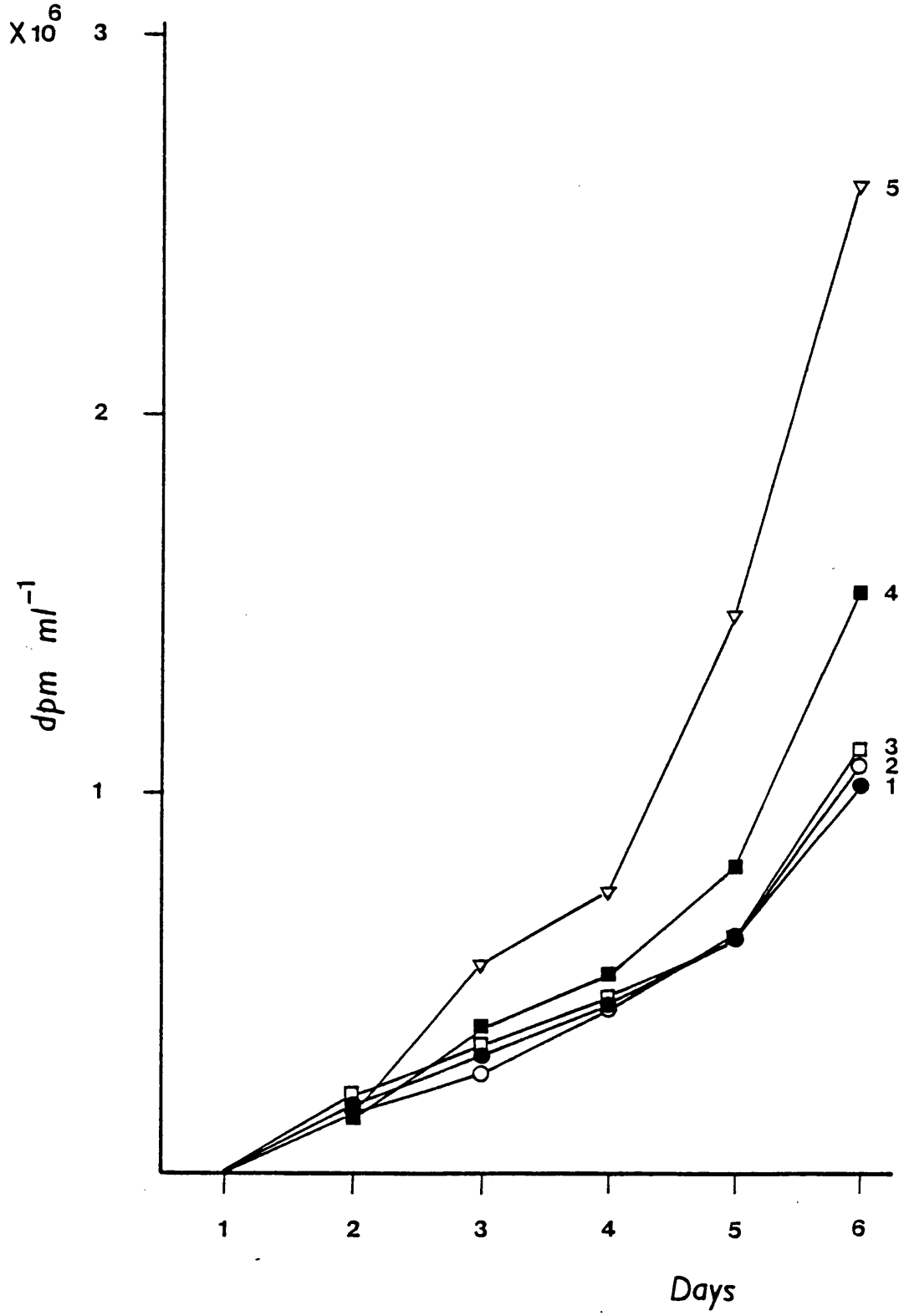

X 10^6
3
2
1
dpm ml^-1
5
4
3
2
1
1
2
3
4
5
6
Days

Figure 7.14 ^{14}C-fixation by nitrogen-starved cells of *Thalassiosira sp.*

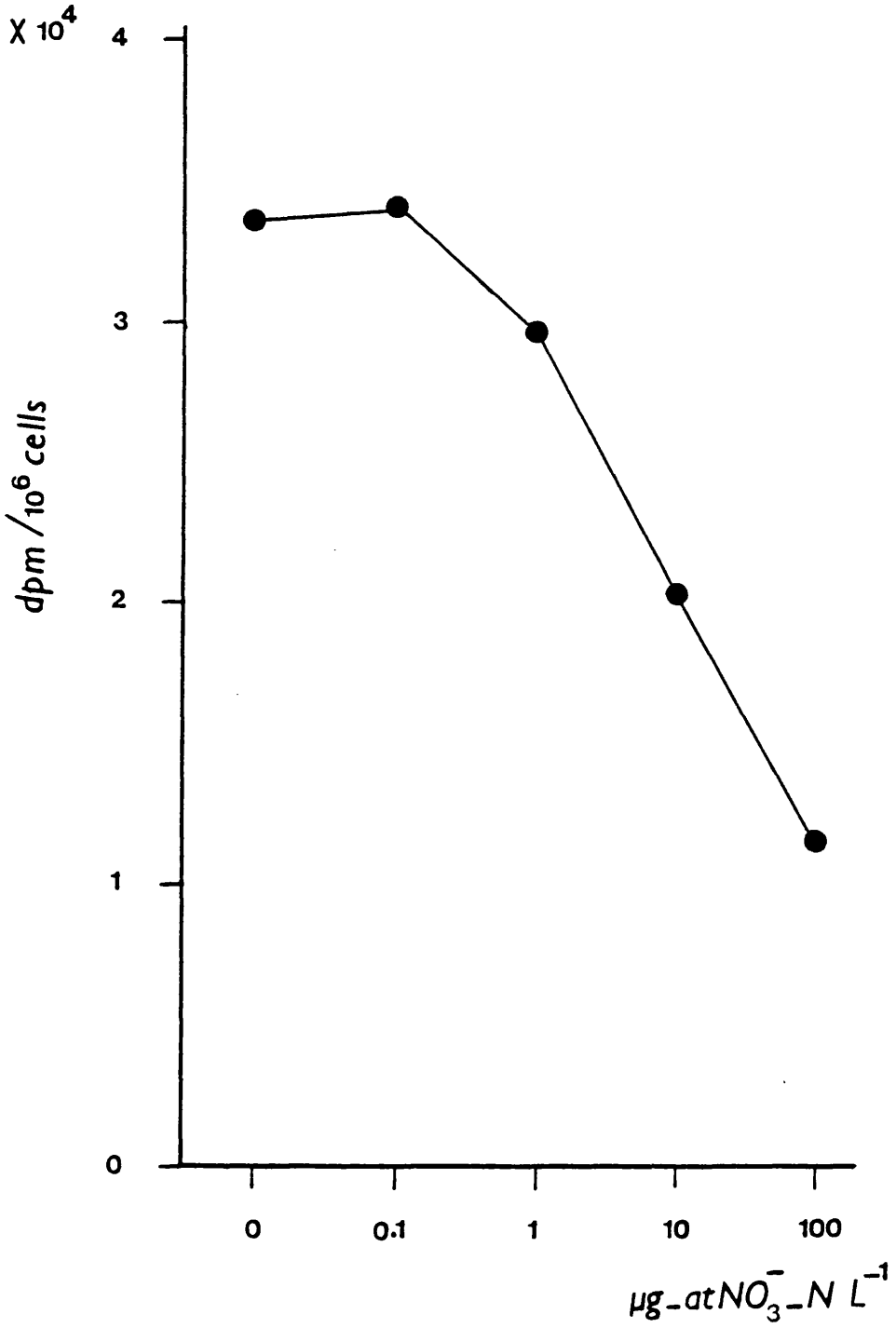
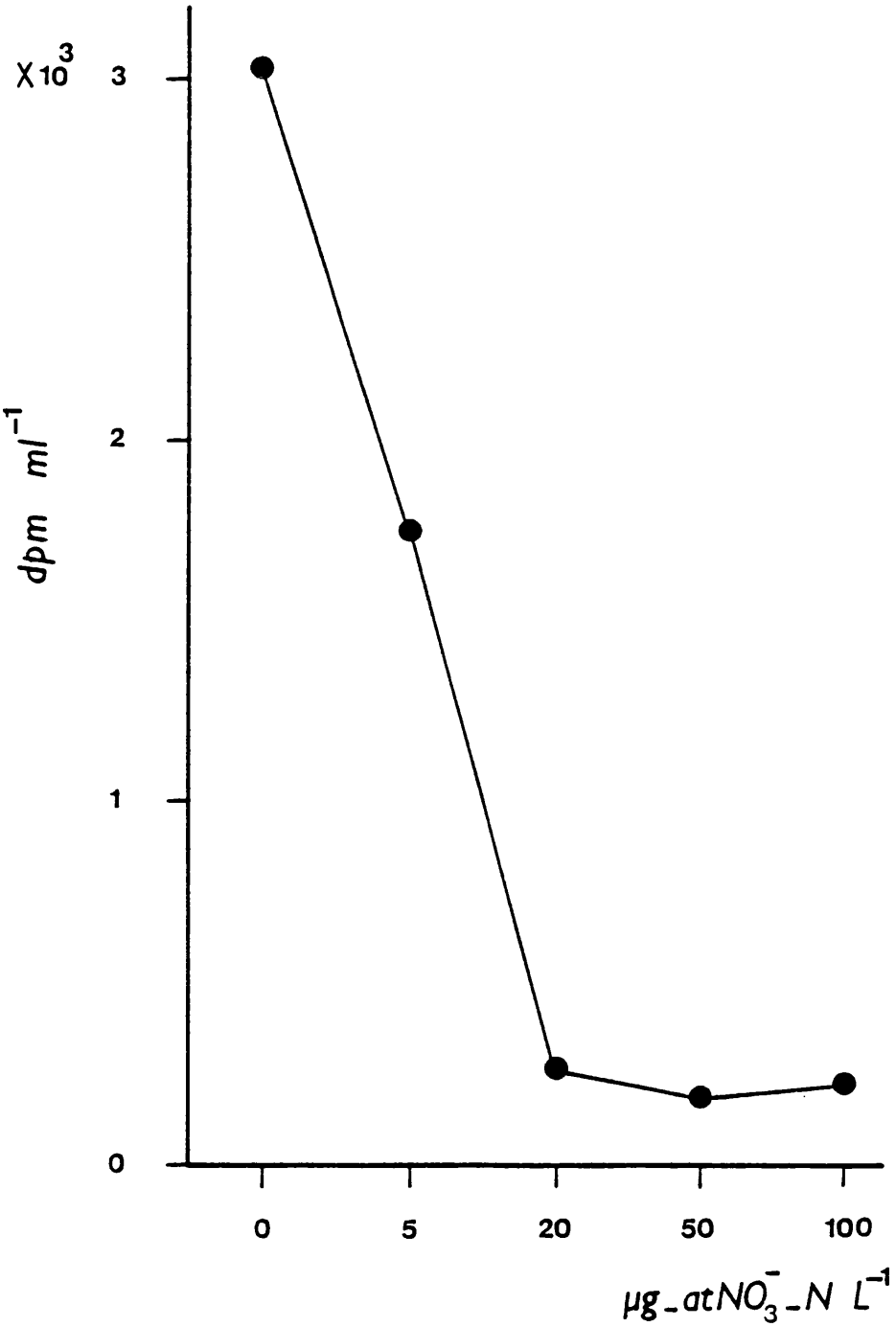

X 10^4
4
3
2
1
0
dpm /10^6 cells
0
0.1
1
10
100
µg-at NO3- N L-1

Figure 7.15 ^{14}C-fixation by nitrogen-starved cells of *Asterionella japonica*


X 10^3
3
2
1
0
dpm ml^-1
0
5
20
50
100
μg_at NO3-_N L^-1

iii. Effect of nitrate on ^{14}C-fixation by Thalassiosira sp. at different time intervals (Fig. 7.16):

In Figure 7.16 the ^{14}C-fixation is plotted against time. At zero time, the ^{14}C-fixation by Thalassiosira cells in the control was 0.163×10^4 dpm/10^6 cells. This value has increased rapidly with time and it reached 4.662×10^4 dpm/10^6 cells after 4 hours of incubation. The ^{14}C-fixation by cells at 1 µg at Nl^{-1} was 0.219×10^4 dpm/10^6 cells at zero time. It reached 1.7×10^4 dpm/10^6 cells after 4 hours of incubation. ^{14}C-fixation by cells at 10 µg at Nl^{-1} increased from 0.051×10^4 dpm/10^6 cells at zero time to 2.0×10^4 dpm/10^6 cells after 4 hours. The ^{14}C-fixation by cells at 100 µg at Nl^{-1} increased from 0.086×10^4 dpm/10^6 cells at zero time to 2.435×10^4 dpm/10^6 cells after 4 hours of incubation. And finally, the ^{14}C-fixation by Thalassiosira cells at 1000 µg at Nl^{-1} increased from 0.061×10^4 dpm/10^6 cells at zero time to 2.669×10^4 dpm/10^6 cells after 4 hours of incubation.

iv. ^{14}C-fixation by Thalassiosira sp. cells previously adapted to different nitrate concentrations (Fig. 7.17):

Thalassiosira cells previously adapted to nitrate for 24 hours have shown an increase of ^{14}C-fixation with higher concentration of nitrate. After 4 hours of incubation, the value of ^{14}C-fixation at 0 µg at l^{-1} was 0.63×10^6 dpm/10^6 cells. At 1 µg at l^{-1}, the ^{14}C-fixation was 0.693×10^6 dpm/10^6 cells. At 10 µg at l^{-1} it was 1.387×10^6 dpm/10^6 cells. At 100 µg at l^{-1} it was 2.162×10^6 dpm/10^6 cells. The ^{14}C-fixation at 1000 µg at l^{-1} was 2.15×10^6 dpm/10^6 cells.

Figure 7.16 ^{14}C-fixation by nitrogen-starved Thalassiosira sp.

1. control
2. 1 µg at Nl^{-1}
3. 10 µg at Nl^{-1}
4. 100 µg at Nl^{-1}
5. 1000 µg at Nl^{-1}

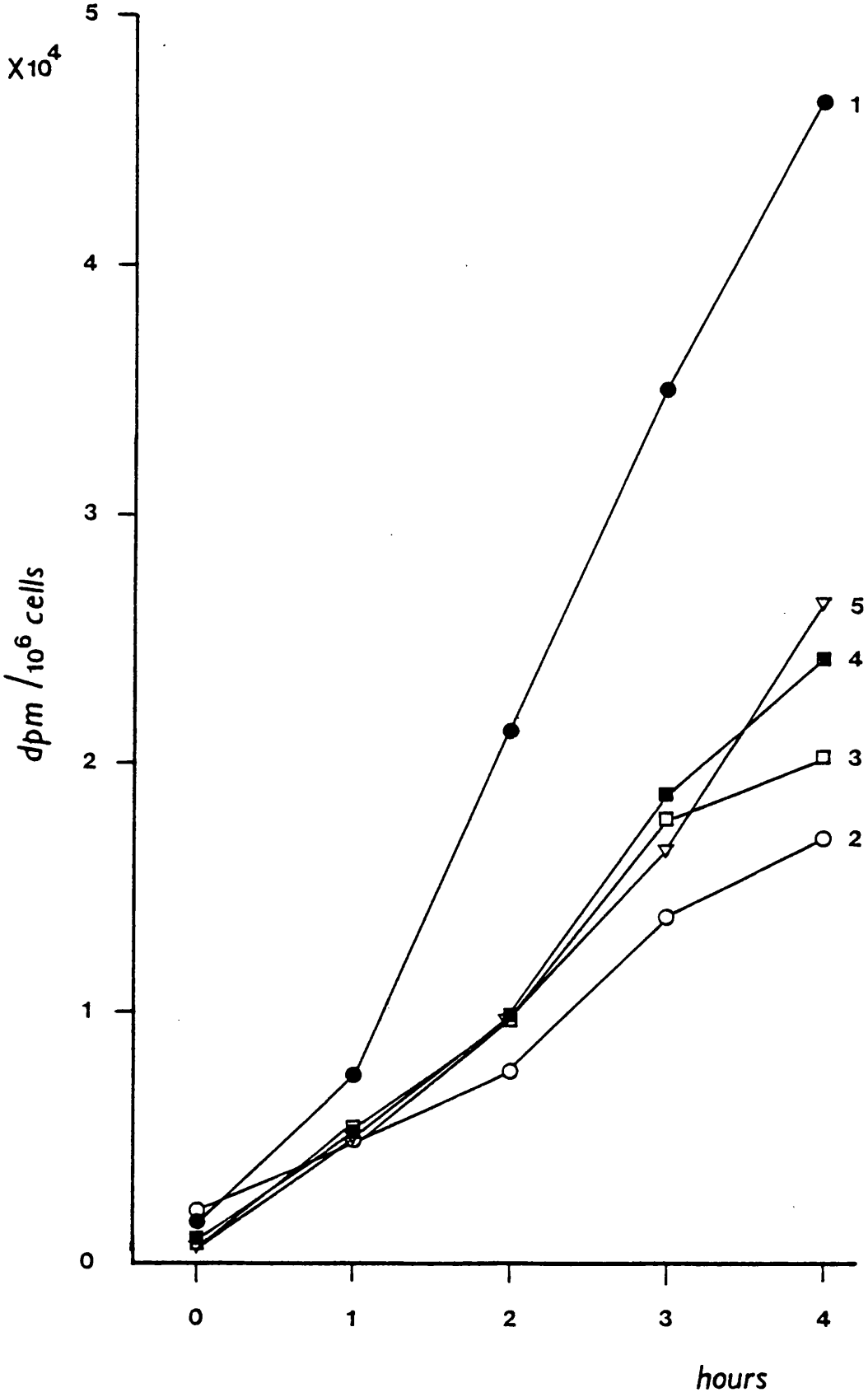
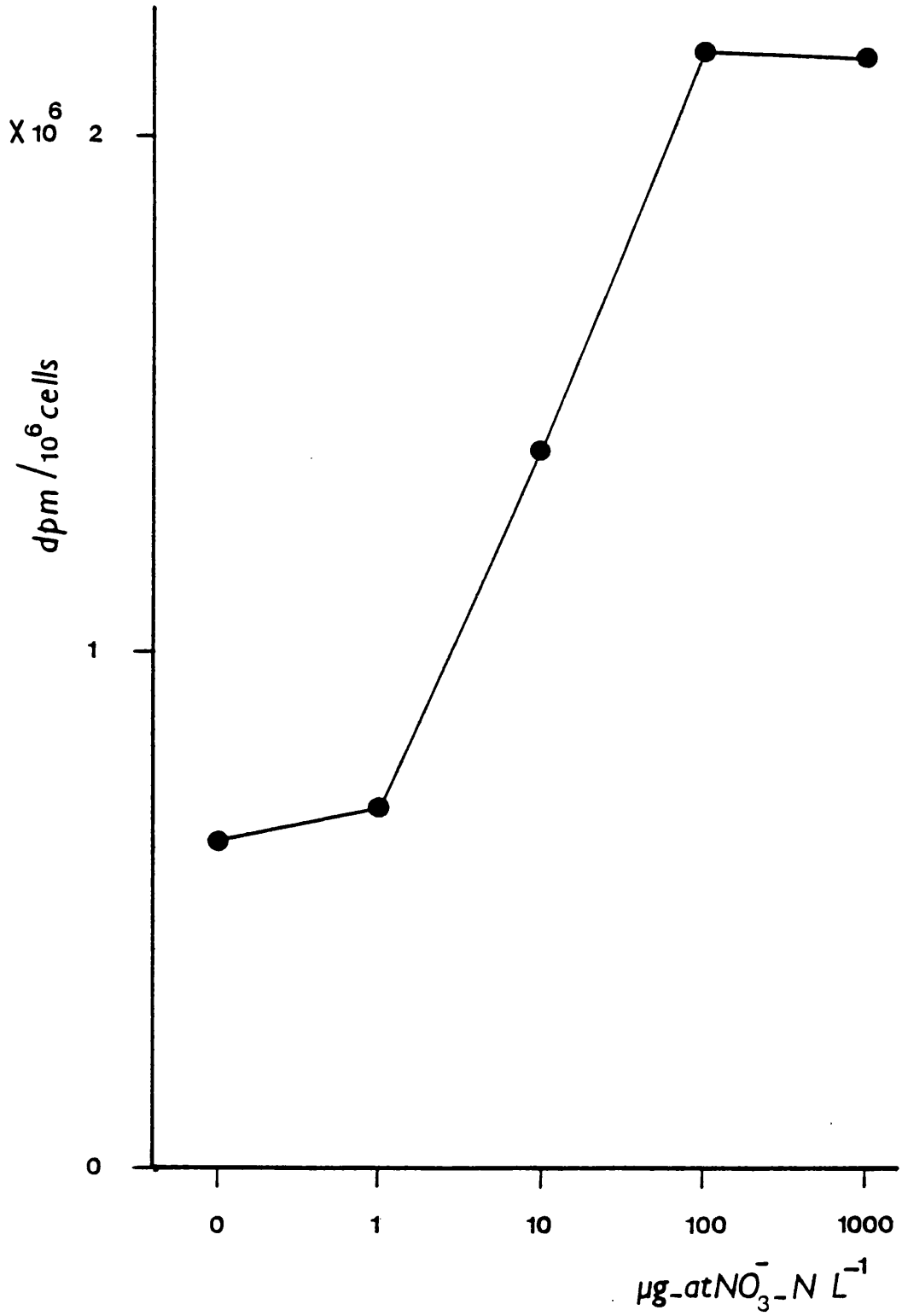

X10^4
5
4
3
2
1
0
dpm /10^6 cells
0
1
2
3
4
hours
1
5
4
3
2

Figure 7.17 ^{14}C-fixation by Thalassiosira sp. previously adapted to nitrate


X 10^6
2
1
0
dpm /10^6 cells
0
1
10
100
1000
µg-at NO3- N L-1

v. Effect of nitrate on the chlorophyll a content of N-starved Thalassiosira sp. (Fig. 7.18):

During a relatively short experiment (72 hours), chlorophyll a content of Thalassiosira cells grown at 500 µg at Nl^{-1} increased rapidly from 0.9 mg chl.a/10^6 cells at zero time up to 5.83 mg chl.a/10^6 cells after 72 hours. At 5 g at Nl^{-1}, chlorophyll a content increased from 0.9 mg chl.a/10^6 cells to 1.4 mg chl.a/10^6 cells after 72 hours. After 72 hours of incubation, chlorophyll a content at 0 µg at Nl^{-1} increased from 0.9 mg chl.a/10^6 cells to 1.13 mg chl.a/10^6 cells.

III. Seawater enrichment and the ^{14}C-fixation by phytoplankton size fractions:

The ^{14}C-fixation values in each of the following experiments are expressed as percentages of the highest ^{14}C-fixation value of that particular experiment.

a. Experiment 1:

Size class 0.45 - 5 µm (Fig. 7.19):

^{14}C-fixation by this size class was stimulated by the addition of phosphate or nitrate. The highest values were found after two days of enrichment. In the control, the highest ^{14}C-fixation percentage was 11%. It increased to 16.9% in culture enriched with 0.5 µg at Pl^{-1}, 22.1% in cultures enriched with 10 µg at Pl^{-1}, 20.7% in cultures enriched with 5 µg at Nl^{-1}, and to 25.6% in cultures enriched with 50 µg at l^{-1}. In all the cultures, values of ^{14}C-fixation declined gradually after reaching the maximum.

Size class 5 - 20 µm (Fig. 7.20):

The patterns of ^{14}C-fixation by this size class were different from

Figure 7.18 Effect of nitrate on chlorophyll *a* content of *Thalassiosira sp.*

1. 0 µg at Nl^{-1}
2. +5 µg at Nl^{-1}
3. +500 µg at Nl^{-1}

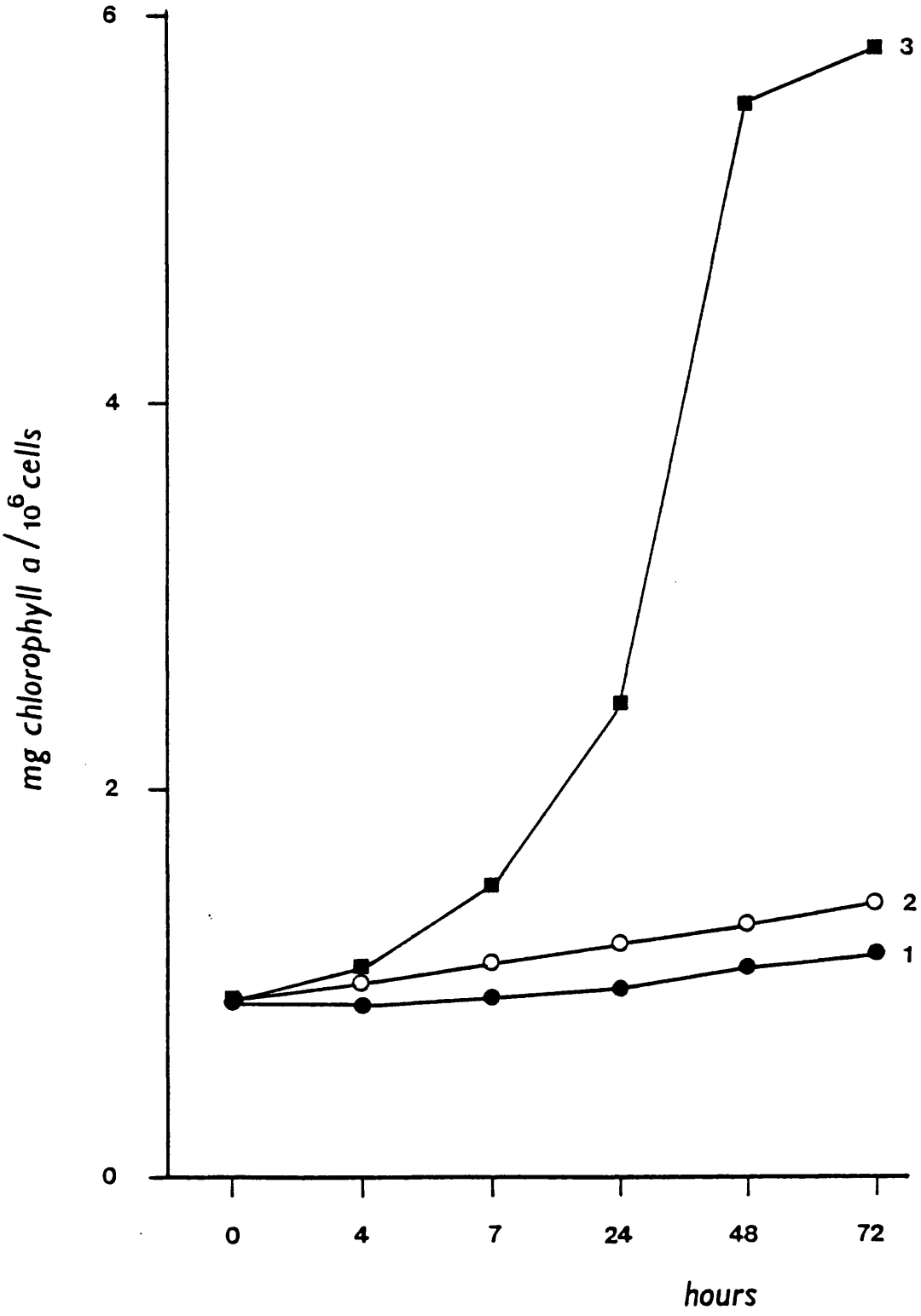

6
4
2
0
mg chlorophyll a / 10^6 cells
0
4
7
24
48
72
hours
3
2
1

Figure 7.19 ^{14}C-fixation by 0.45 - 5 µm size fraction

1. control
2. +0.5 µg at Pl^{-1}
3. +10 µg at Pl^{-1}
4. +5 µg at Nl^{-1}
5. +50 µg at Nl^{-1}

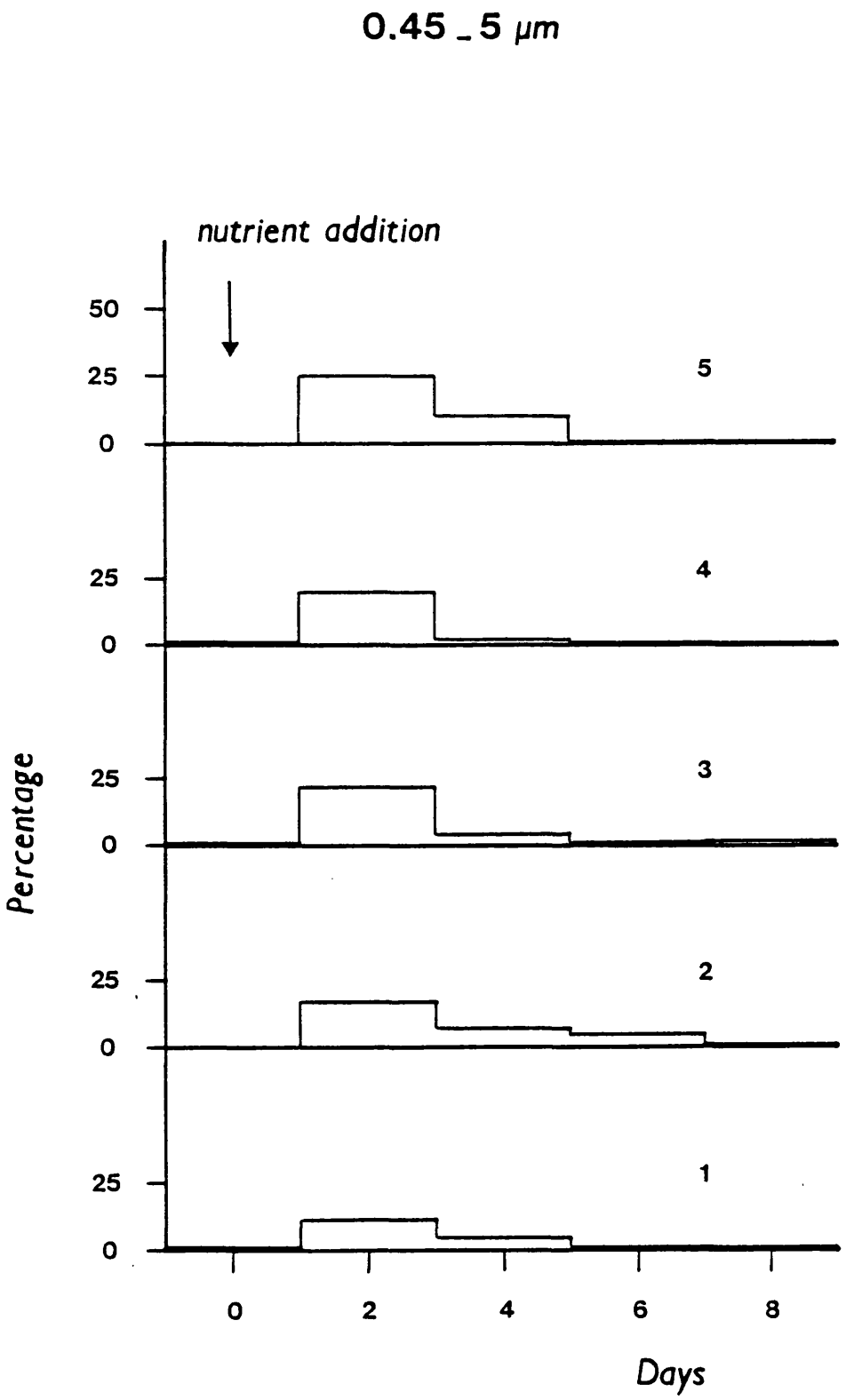
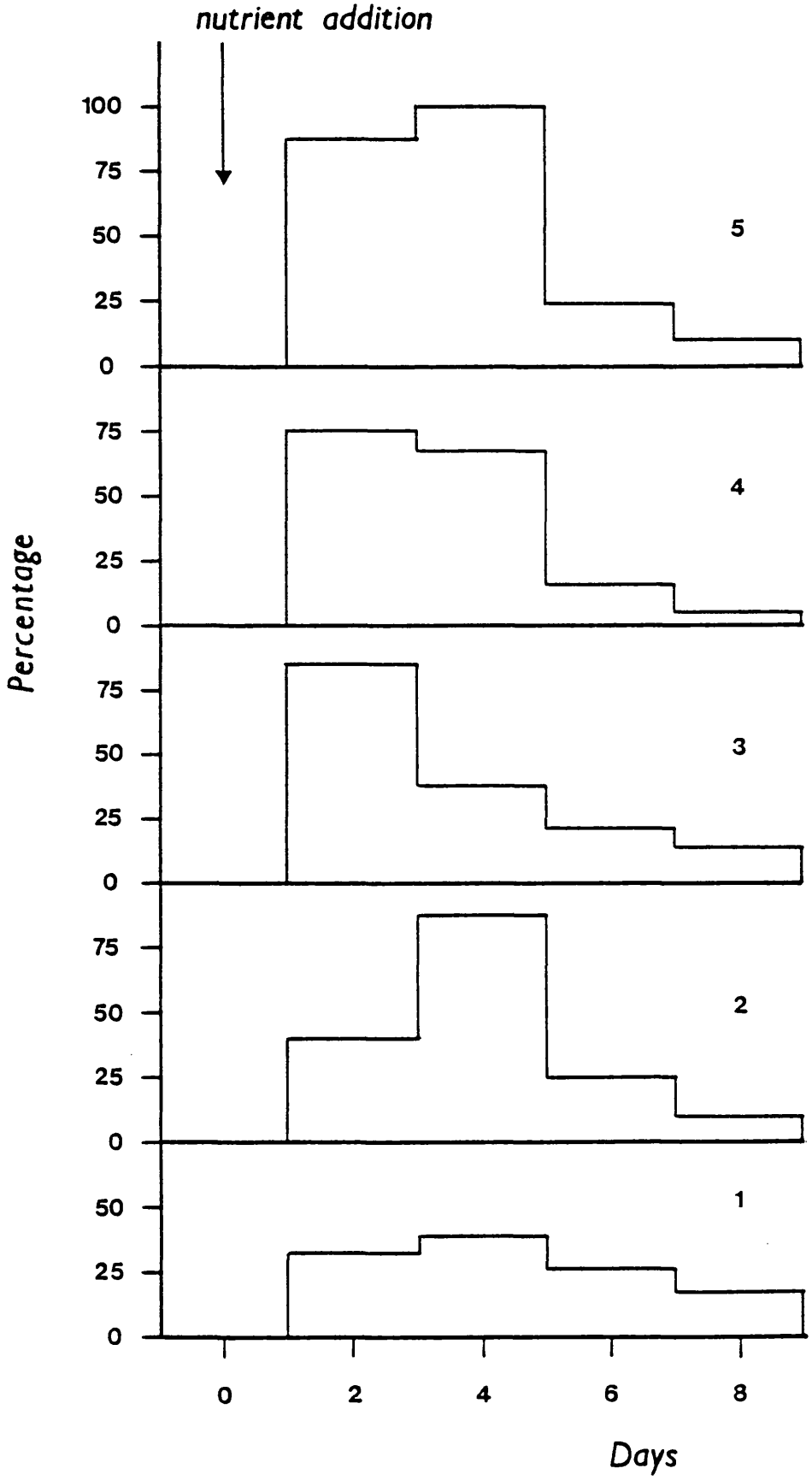

0.45 _ 5 μm
nutrient addition
50
25
0
5
25
0
4
25
0
3
25
0
2
25
0
1
Percentage
0
2
4
6
8
Days

Figure 7.20 ^{14}C-fixation by 5 - 20 µm size fraction

1. control
2. +0.5 µg at $P1^{-1}$
3. +10 µg at $P1^{-1}$
4. +5 µg at $N1^{-1}$
5. +50 µg at $N1^{-1}$

5 _ 20 μm


nutrient addition
100
75
50
25
0
5
75
50
25
0
4
Percentage
75
50
25
0
3
75
50
25
0
2
50
25
0
1
0
2
4
6
8
Days

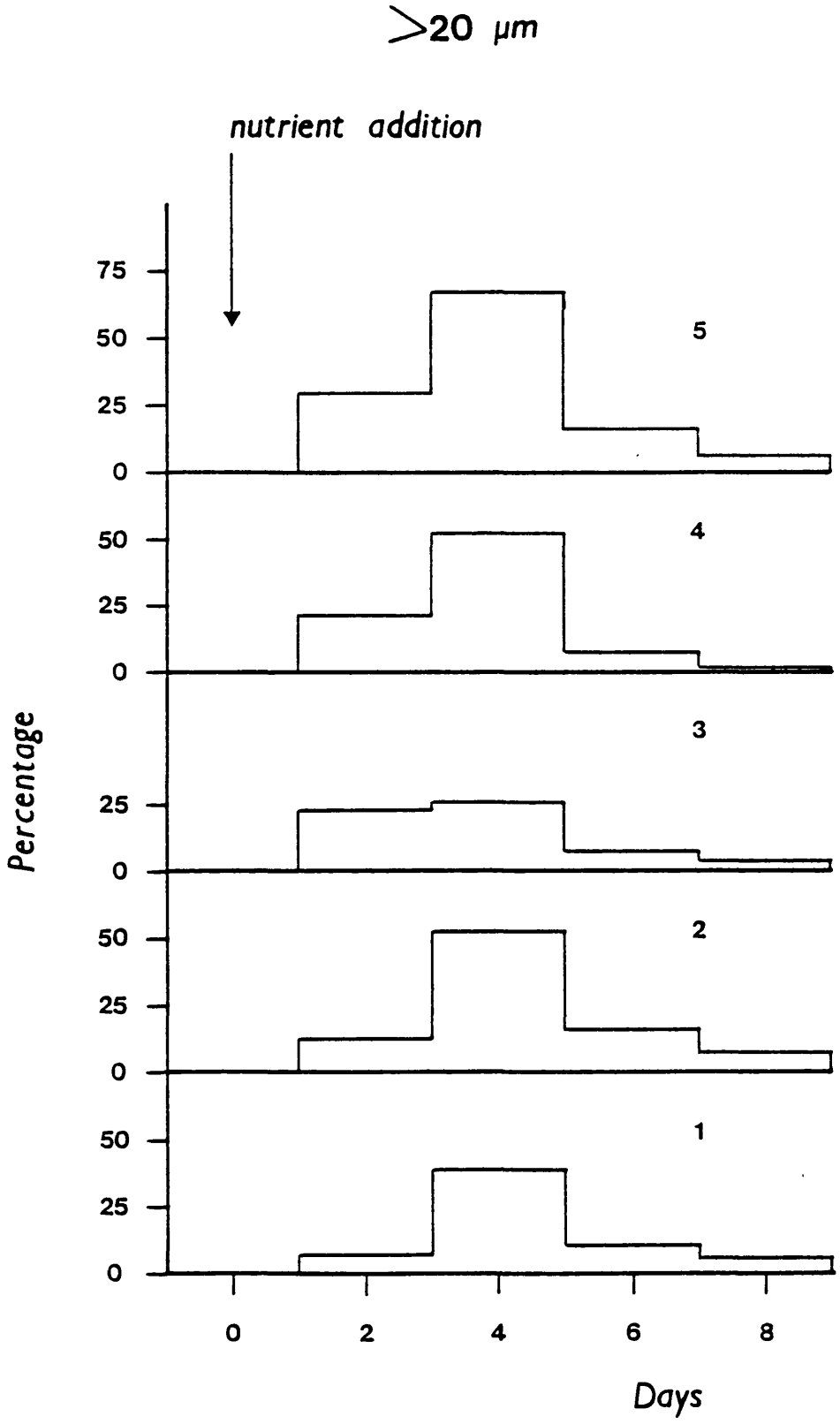
those of 0.45 - 5 μm size class. In all the cultures there was an increase in ^{14}C-fixation as a result of nutrient enrichment. In the culture enriched with 0.5 μg at $P1^{-1}$, ^{14}C-fixation increased significantly after two days, but reached its maximum of 87.2% after four days of enrichment. It dropped to 24.8% on the sixth day and down to 9.8% after eight days of incubation. Culture enriched with 10 μg at $P1^{-1}$ reached its maximum after two days of incubation reaching 85.8%. This was followed by a gradual decrease down to 14.1%. In the culture enriched with 5 μg at $N1^{-1}$, the ^{14}C-fixation reached its highest values on the second and fourth days of incubation (75.7%, 68.1% respectively). The value then reached 5.7% on the eighth day. ^{14}C-fixation pattern in culture enriched with 50 μg at $N1^{-1}$ was similar to that of culture enriched with 5 μg at $N1^{-1}$. A high value of 87.2% was found on the second day followed by a maximum value of 100% on the sixth day. This was followed by a rapid decrease down to 10.9% on the eighth day.

Size class >20 μm (Fig. 7.21):

In all the cultures, the highest ^{14}C-fixation by phytoplankton >20 μm occurred on the fourth day of incubation. In the control, the maximum ^{14}C-fixation was 39.8%. The value in culture enriched with 0.5 μg at $P1^{-1}$ reached 52.8% on the fourth day. In the culture enriched with 10 μg at $P1^{-1}$ the fixation value reached 22.9% on the second day and 25.6% on the fourth day. Maximum value of ^{14}C-fixation in the culture enriched with 5 μg at $N1^{-1}$ was 53.4% on the fourth day, while the maximum value of culture enriched with 50 μg at $N1^{-1}$ was 68.1% on the fourth day. In all the cultures, including the control, the ^{14}C-fixation values decreased after the fourth day until they reached their lowest values on the eighth day.

Figure 7.21 ^{14}C-fixation by > 20 µm size fraction

1. control
2. +0.5 µg at Pl^{-1}
3. +10 µg at Pl^{-1}
4. +5 µg at Nl^{-1}
5. +50 µg at Nl^{-1}


>20 μm
nutrient addition
75
50
25
0
5
50
25
0
4
25
0
3
50
25
0
2
50
25
0
1
Percentage
0
2
4
6
8
Days

b. Experiment 2:

Size class 0.45 - 5 µm (Fig. 7.22):

^{14}C-fixation by phytoplankton in the control increased slightly to 7.4% on the third day of the incubation. The fixation in the culture enriched with 50 µg at Nl^{-1} increased to its maximum value of 35.1% on the sixth day of the incubation. Highest ^{14}C-fixation values in the culture enriched with 500 µg at Nl^{-1} were 14.8% on the sixth day and 18.9% on the ninth day. This was then followed by a steady decrease down to 3.4% on the sixteenth day of the experiment.

Size class 5 - 20 µm (Fig. 7.23):

^{14}C-fixation by phytoplankton in the control increased to 16.5% on the third day and up to 19.6% on the sixth day. This was followed by a steady decrease down to 1.4% on the sixteenth day (Fig. 7.23.1). In the culture enriched with 50 µg at Nl^{-1}, the ^{14}C-fixation value increased to 27% on the third day and then to 64.8% on the sixth day. On the ninth day, ^{14}C-fixation dropped to 9.8% and remained at this level until the end of the experiment (Fig. 7.23.2). ^{14}C-fixation in the culture enriched with 500 µg at Nl^{-1} increased from 0.2% at the start of the experiment and before the enrichment to 10.1% on the third day as a result of nutrient enrichment. This value increased to 27.7% on the sixth day, but decreased slightly to 15.5% on the ninth day. A very high ^{14}C-fixation value was found on the thirteenth day (100%) followed by a drop to 47.3% at the end of the experiment.

Size class >20 µm (Fig. 7.24):

In the control, ^{14}C-fixation value increased very slightly to a maximum value of 8.1% on the sixth day. In the culture enriched with

Figure 7.22 ^{14}C-fixation by 0.45 - 5 µm size fraction

1. control
2. +50 µg at Nl^{-1}
3. +500 µg at Nl^{-1}

0.45 _ 5 μm

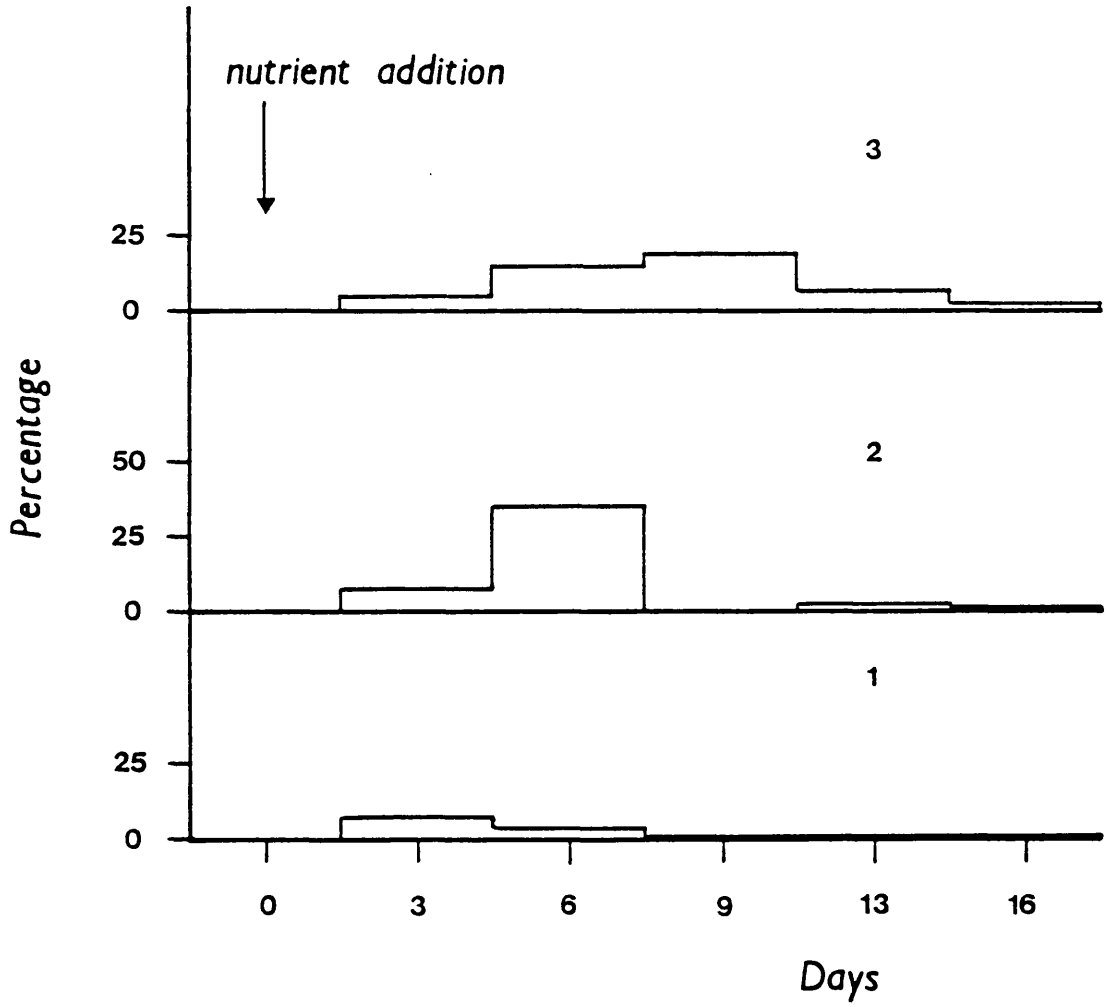

nutrient addition
3
25
0
2
50
25
0
1
25
0
Percentage
0
3
6
9
13
16
Days

Figure 7.23 ^{14}C-fixation by 5 - 20 µm size fraction

1. control
2. +50 µg at Nl^{-1}
3. +500 µg at Nl^{-1}

5_20 μm

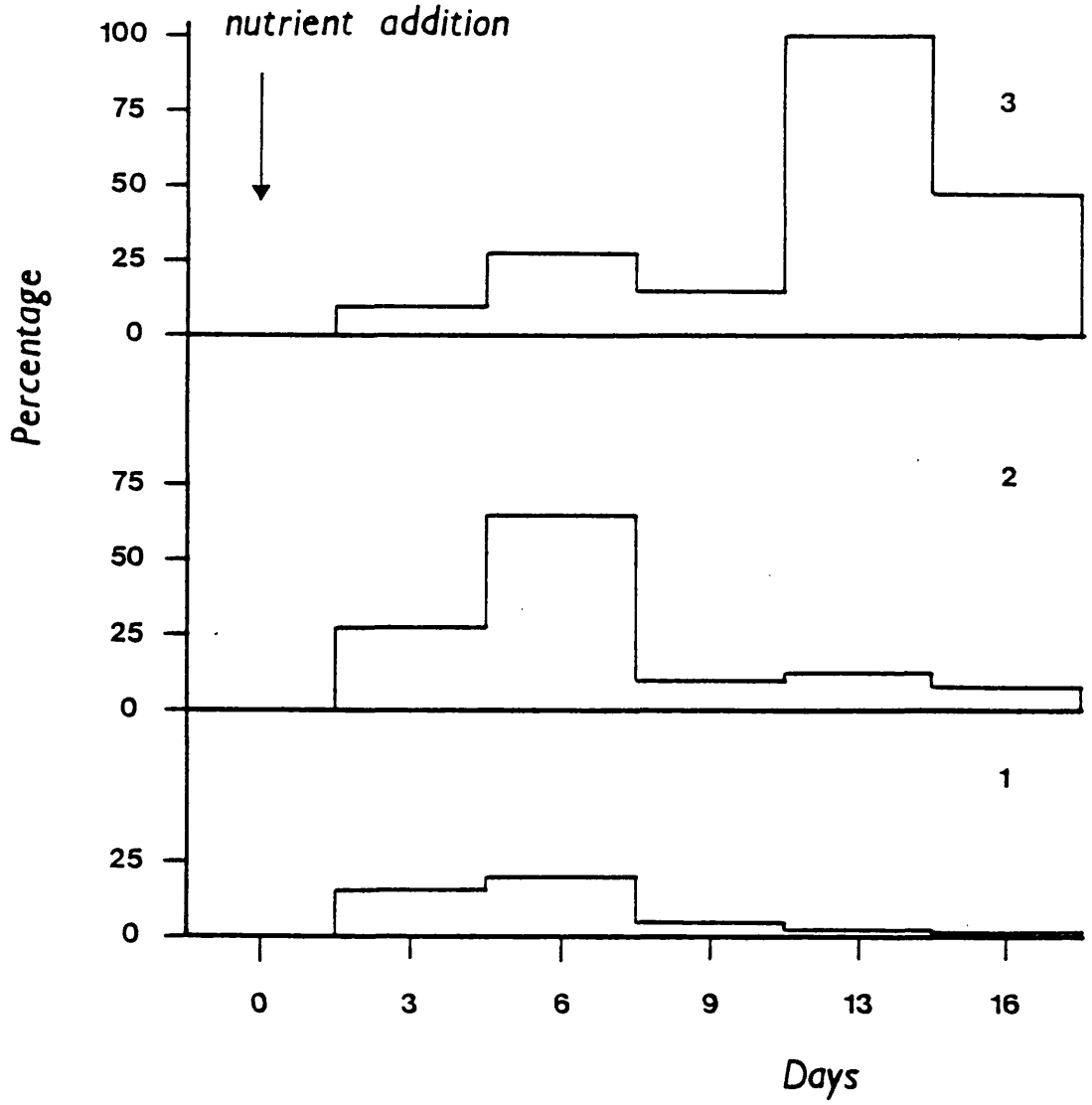
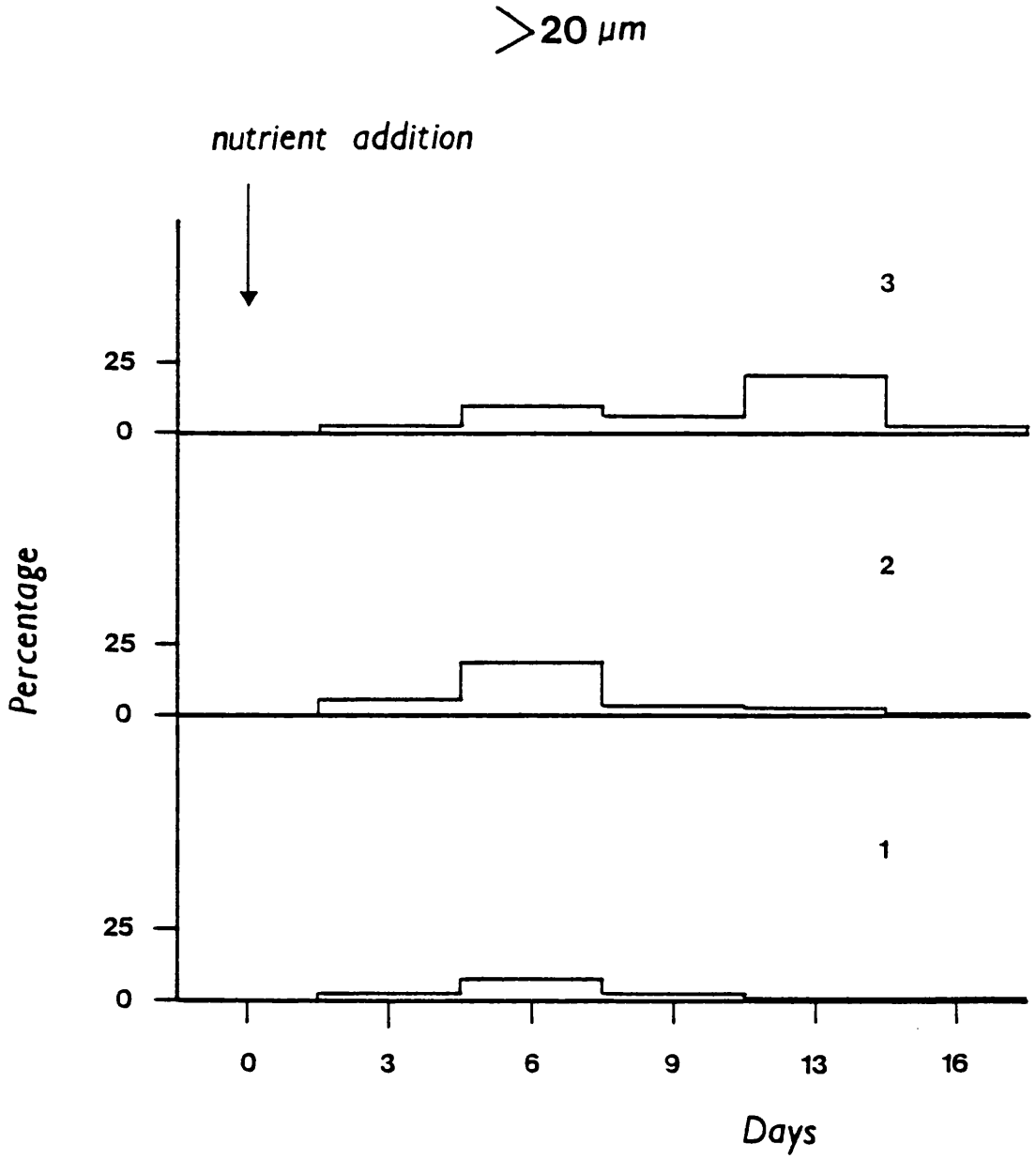

nutrient addition
100
75
50
25
0
3
75
50
25
0
2
25
0
1
Percentage
0
3
6
9
13
16
Days

Figure 7.24 ^{14}C-fixation by >20 µm size fraction

1. control
2. +50 µg at Nl^{-1}
3. +500 µg at Nl^{-1}


>20 µm
nutrient addition
3
2
1
25
0
Percentage
0
3
6
9
13
16
Days

50 µg at Nl^{-1}, the ^{14}C-fixation reached its maximum value of 19.2% on the sixth day. This was followed by a steady decrease down to 3% on the sixteenth day. From the third day to the ninth day of the experiment, the ^{14}C-fixation value by cells enriched with 500 µg at l^{-1} was around 6%. This value increased to 20.9% on the thirteenth day and then dropped to 2.7% on the sixteenth day.

c. Experiment 3:

Nitrate uptake (Fig. 7.25):

Nitrate concentration in the control decreased steadily from 35 µg at Nl^{-1} at the start of the experiment down to undetectable concentration on the sixth day of the experiment. Nitrate concentration in the culture enriched with 50 µg at Nl^{-1} decreased steadily from 85 µg at Nl^{-1} at the beginning of the experiment to undetectable concentration on the sixth day of the experiment. Nitrate concentration in culture enriched with 500 µg at Nl^{-1} decreased steadily from 535 µg at Nl^{-1} at the beginning of the experiment to 518 µg at Nl^{-1} on the fourth day. Thereafter, there was a rapid decrease in nitrate concentration until it reached 183 µg at Nl^{-1} on the last day of the experiment.

Size class 0.45 - 5 µm (Fig. 7.26):

In the control, ^{14}C-fixation reached its highest value on the sixth day of the experiment (11.9%), but decreased afterwards. In the culture enriched with 50 µg at Nl^{-1}, the highest ^{14}C-fixation value was found on the sixth day (19.4%) of the experiment followed by a gradual decrease down to 4% on the tenth day. ^{14}C-fixation in the culture enriched with 500 µg at Nl^{-1} increased gradually up to 21.2% on the eighth day of the experiment. This was followed by a drop down to 6.6%

Figure 7.25 Nitrate uptake by phytoplankton measured as loss from medium

a. control
b. +50 µg at Nl^{-1}
c. +500 µg at Nl^{-1}

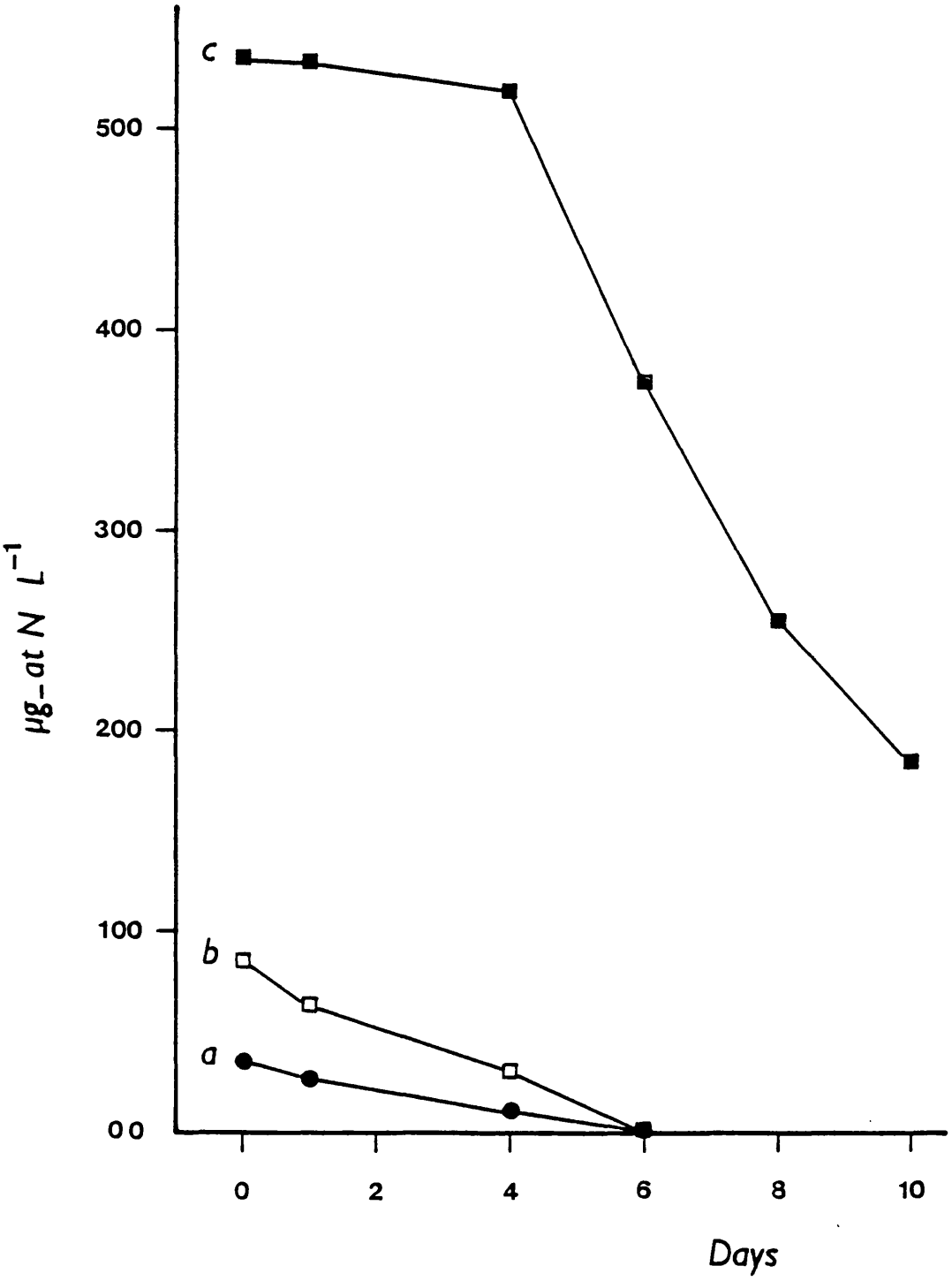
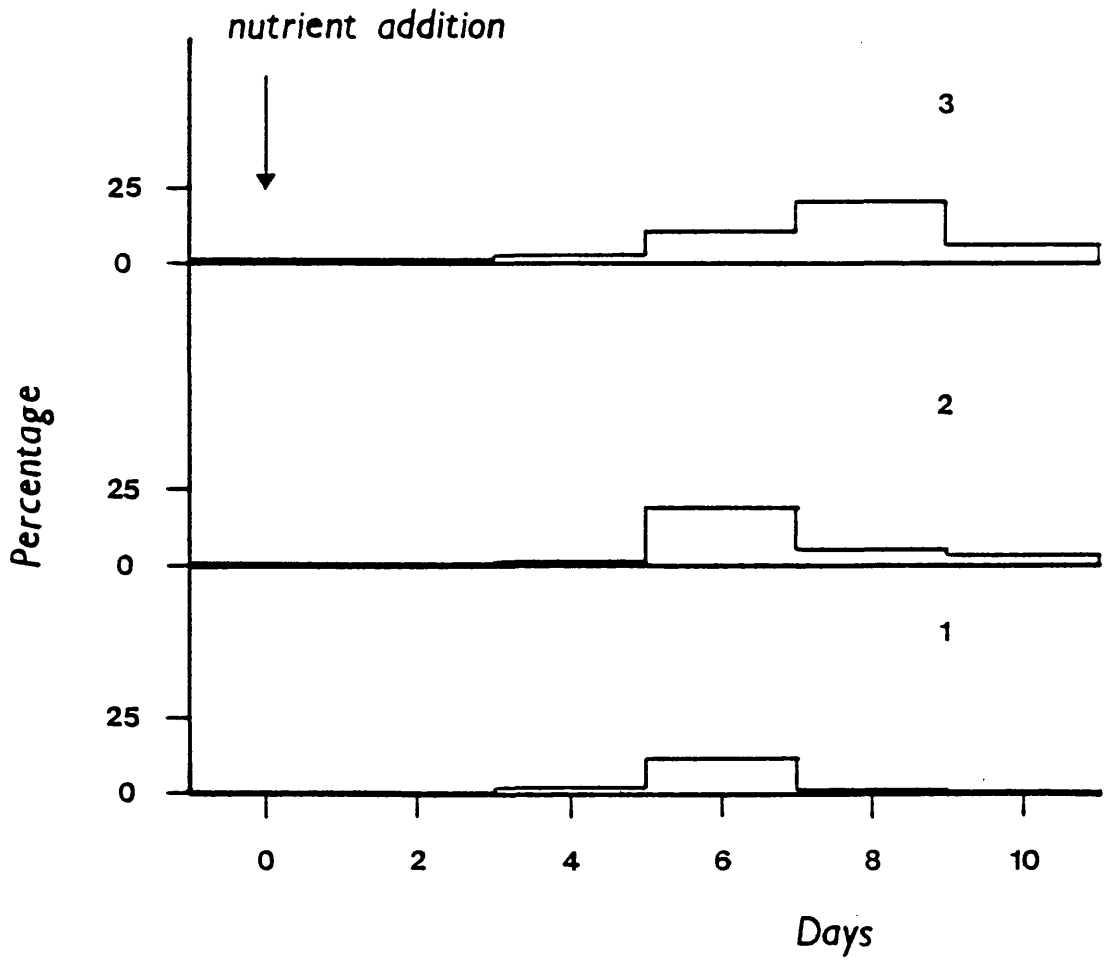

c
500
400
300
200
100
b
a
00
0
2
4
6
8
10
Days
µg_at N L⁻¹

Figure 7.26 ^{14}C-fixation by 0.45 - 5 µm size fraction

1. control
2. +50 µg at Nl^{-1}
3. +500 µg at Nl^{-1}


0.45_5 μm
nutrient addition
3
25
0
2
25
0
1
25
0
Percentage
0
2
4
6
8
10
Days

on the tenth day.

Size class 5 - 20 µm (Fig. 7.27):

In the control culture, ^{14}C-fixation increased to 21.2% on the fourth day and up to 58.1% on the sixth day. This was followed by a sudden drop down to 6% on the eighth day (Fig. 7.27.1). In the culture enriched with 50 µg at Nl^{-1}, the ^{14}C-fixation value increased from 1.0% on the second day, to 38.9% on the fourth day, and up to 100% on the sixth day of the experiment. Then it dropped to 16.7% on the eighth day (Fig. 7.27.2). ^{14}C-fixation in the culture enriched with 500 µg at Nl^{-1} increased from 29.8% on the fourth day to 100% on the sixth day of the experiment. This value dropped from 100% to 46.5% and down to 25.7% on the tenth day (Fig. 7.27.3)

Size class >20 µm (Fig. 7.28):

In the control culture, ^{14}C-fixation reached its highest value of 27.8% on the sixth day of the experiment. In the culture enriched with 50 µg at Nl^{-1}, ^{14}C-fixation increased from 1.0% on the second day, to 14.1% on the fourth day, and up to 49.7% on the sixth day. This was followed by a decrease to 10.1% on the eighth day, and down to 4.0% on the last day of the experiment. The ^{14}C-fixation pattern in the culture enriched with 500 µg at Nl^{-1} was slightly different from those in the control, and in the culture enriched with 50 µg at Nl^{-1}. The ^{14}C-fixation increased from 0.5% on the second day to 15.6% on the fourth day and up to 46.7% on the sixth day of the experiment. This was followed by a drop down to 16.2% on the eighth day and then, an increase up to 39.4% on the last day of the experiment.

Figure 7.27 ^{14}C-fixation by 5 - 20 µm size fraction

1. control
2. +50 µg at Nl^{-1}
3. +500 µg at Nl^{-1}

5_20 μm

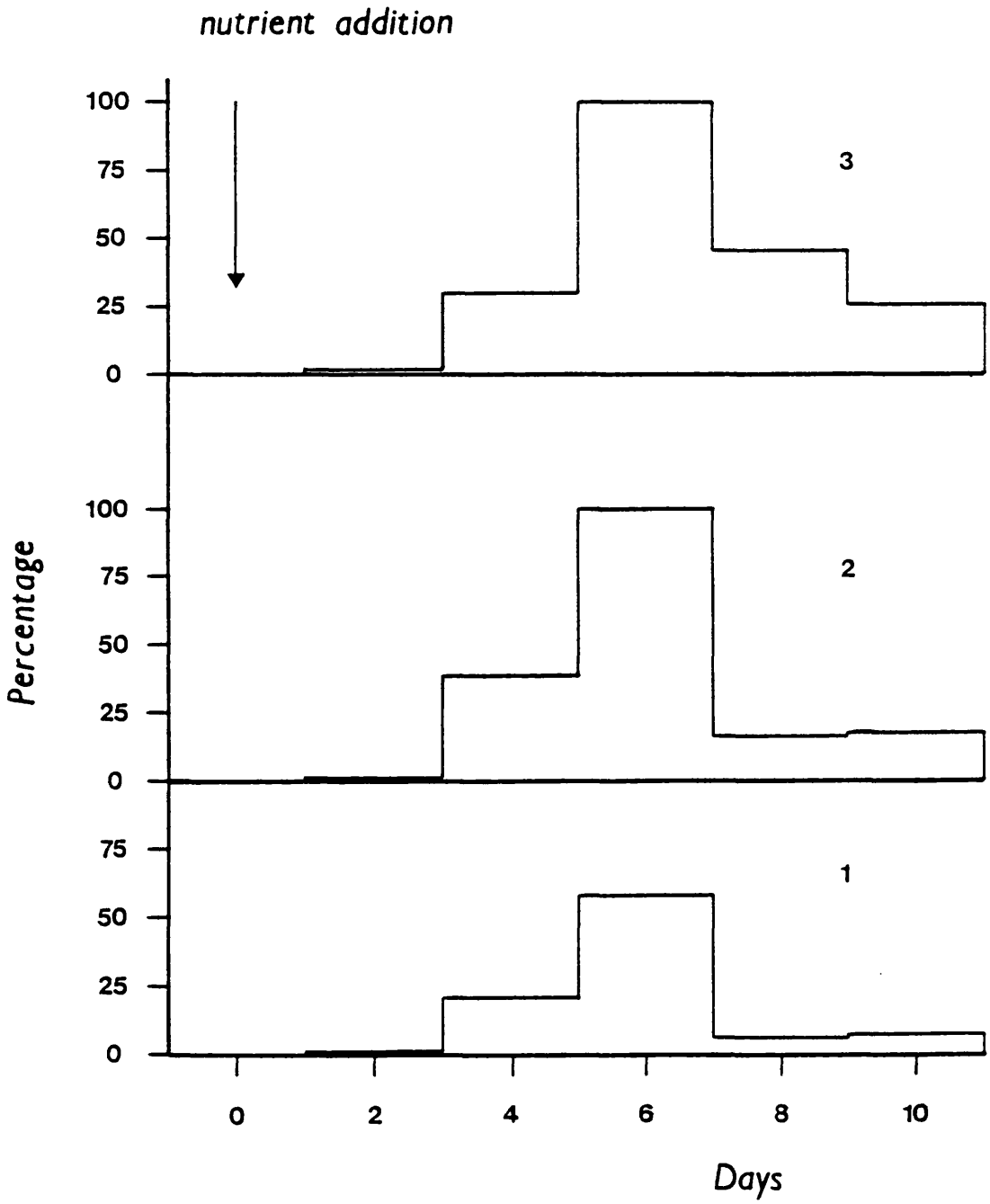
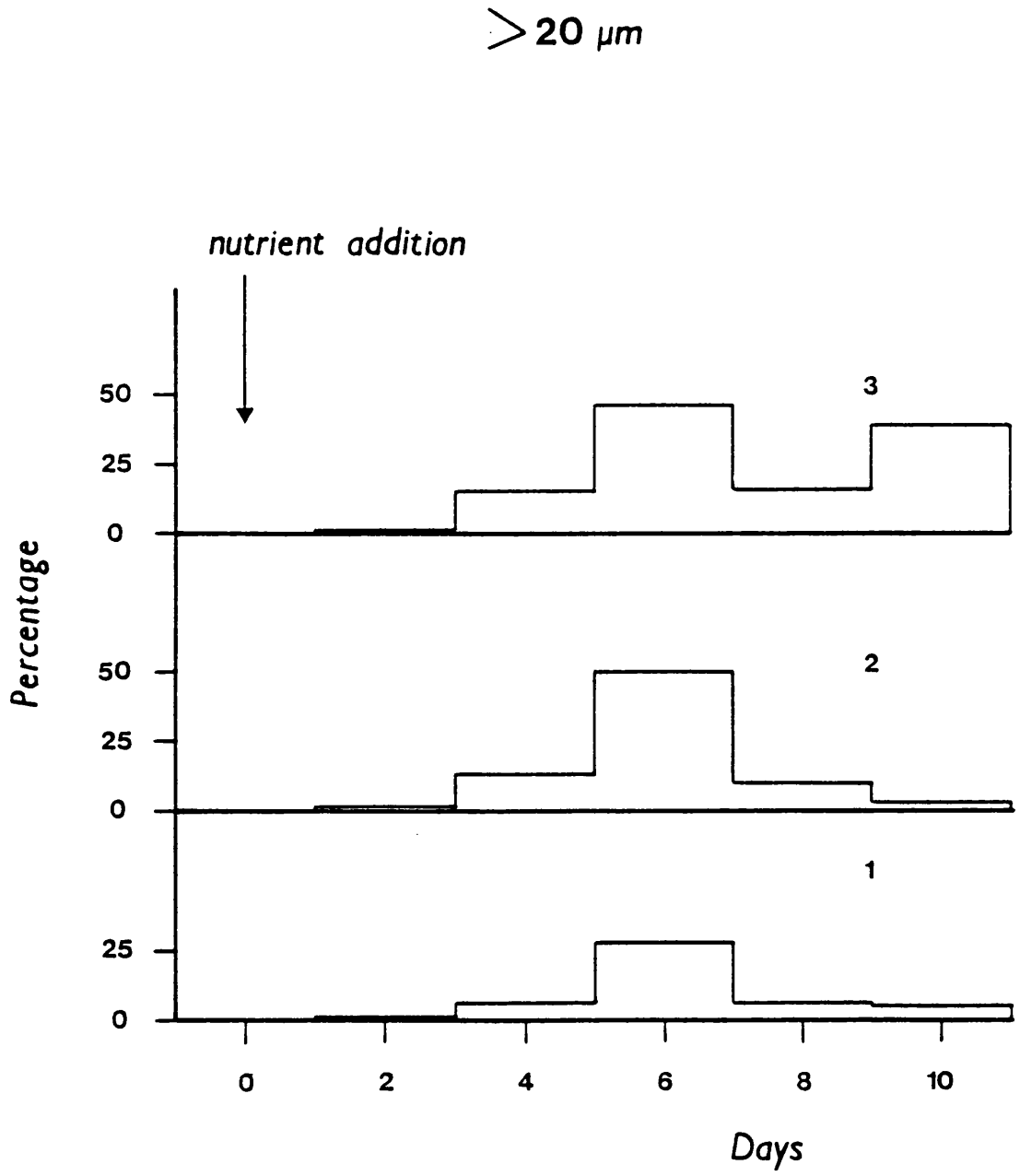

nutrient addition
100
75
50
25
0
3
100
75
50
25
0
2
75
50
25
0
1
Percentage
0
2
4
6
8
10
Days

Figure 7.28 ^{14}C-fixation by >20 µm size fraction

1. control
2. +50 µg at Nl^{-1}
3. +500 µg at Nl^{-1}


> 20 μm
nutrient addition
Percentage
50
25
0
3
50
25
0
2
25
0
1
0
2
4
6
8
10
Days

DISCUSSION

A universal artificial medium which can be used for physiological experiments of phytoplankton was and still is an objective hard to achieve. Figures 7.2 - 7.4 show the results of experiments with a modified version of Aquil medium (Morel et al., 1979). Although Aquil medium did not match Erdschreiber medium as a growth medium, nevertheless the three phytoplankton species tested showed a satisfactory growth. ^{14}C-fixation has been used in this study as an indicator of the growth of phytoplankton wherever possible. Figure 7.5 a, b shows the ^{14}C-fixation by Thalassiosira sp., Prorocentrum micans and Ditylum brightwellii in unialgal cultures. When ^{14}C-fixation was expressed per 10^6 cells of each species (Fig. 7.5 a), D. brightwellii had the highest value followed by P. micans and then Thalassiosira sp.. This order of fixation corresponded to the decrease in surface area from D. brightwellii (11314 μm^2), P. micans (1540 μm^2) to Thalassiosira sp. (707 μm^2). But when ^{14}C-fixation was expressed per $\mu m^2.10^6$ cells (Fig. 7.5 b), P. micans had the highest value followed by Thalassiosira sp., and then D. brightwellii. This order agrees with the point of view which stresses that cells with the highest surface area to volume ratio and those with more mobility have higher growth rates and hence take up nutrients faster than larger cells with low surface to volume ratios (Williams, 1964; Eppley and Sloan, 1966; Banse, 1976).

Thalassiosira sp., P. micans and D. brightwellii were grown in mixed culture containing full Aquil medium. Thalassiosira sp. had outgrown the other two species when the concentration of cells was considered (Fig. 7.6). This was mainly due to higher growth rate and possibly higher nutrient uptake capacity. D. brightwellii growth was

very low when compared to Thalassiosira sp., which could be the result of lower growth rate due to the comparatively large cell volume. The interesting result was that of P. micans. When this species was grown with Thalassiosira and D. brightwellii it showed very low growth rate. Bearing in mind the smaller cell volume of P. micans when compared with D. brightwellii, the low number of the former species could possibly be the result of an inhibitory extracellular product by Thalassiosira sp.. It seems that P. micans only was affected by this suggested inhibitory product. This is supported by the results of surface area shown in Figure 7.7. From this figure it is obvious that there was a large increase in the total surface area of Thalassiosira sp. and D. brightwellii but not in P. micans.

Figure 7.8 shows the ^{14}C-fixation by the different size fractions in the previous mixed culture experiment. These results show that the highest fixation was carried out by cells between 20-50 μm in diameter. The second highest fixation was carried out by cells between 0.45-20 μm in diameter, and the least fixation was carried out by cells > 50 μm in diameter. This order in ^{14}C-fixation indicates that the highest fixation was carried out by P. micans cells, followed by Thalassiosira sp. cells and then D. brightwellii cells. This result must not be taken entirely at its face value for the fixation by the different species. This is due to the fact that there was a differential retention by each filter. This was confirmed by an experiment conducted to calculate the percentage of Thalassiosira cells which pass through a 20 μm filter, and the percentage of P. micans and D. brightwellii cells which can pass through 50 μm filters. It was found that 90% of Thalassiosira cells pass through the 20 μm filter, and 25% of D. brightwellii cells pass through the 50 μm filter. Using this

result the values in Figure 7.8 were corrected and related to the different species. The ^{14}C-fixation of the different species is illustrated in Figure 7.9. The order of fixation in this figure followed the order observed in Figure 7.6. The ^{14}C-fixation values of the three species in Figure 7.9 are very close to each other. Comparing Figures 7.6 and 7.9 shows that the high fixation value of Thalassiosira was because of its high cell number. The high fixation value of D. brightwellii was because of its high total surface area. Finally the high fixation value of P. micans was because of its high rate of ^{14}C uptake per unit cell number (Fig. 7.10), and high uptake rate per unit surface area (Fig. 7.11). It can be concluded from the mixed culture experiment (Figs. 7.6 - 7.11) that the critical factors in the ^{14}C-fixation by phytoplankton cells are the high growth rate (represented by Thalassiosira) and high uptake capacity per unit surface area (represented by P. micans).

The effect of nitrate on growth of and ^{14}C-fixation by Thalassiosira sp. was investigated and the results are illustrated in Figures 7.12 and 7.13. It can be seen from the results that the increase of nitrate concentration in the medium was accompanied by an increase in cell number. This result emphasizes the role of nitrate as an important nutrient needed by the phytoplankton for growth. This role is further illustrated by an increase of ^{14}C-fixation as a result of an increase in nitrate concentration (Fig. 7.13).

When nitrate was added to nitrogen-starved cells of Asterionella japonica and of Thalassiosira sp., the highest ^{14}C-fixation was carried out by cells in the lowest nitrate concentrations (Figs. 7.14 and 7.15). Since nitrogen-starved cells were in immediate need of nitrate, it was assimilated at a higher rate than was^{14}C-fixation. The very low

values of ^{14}C-fixation by cells supplied with high nitrate concentrations was because energy sources were directed towards nitrate assimilation at the expense of ^{14}C-uptake (Healey, 1973; Falkowski and Stone, 1975; Thomas et al., 1976; Hipkin et al., 1983).

In another experiment, the rate of ^{14}C-fixation was measured in nitrogen-starved Thalassiosira sp. supplied with different nitrate concentrations. The fixation was measured at hourly intervals. At the beginning of the incubation (0 - 1 hour) the energy sources in the phytoplankton supplied with nitrate were directed towards nitrate uptake at the expense of ^{14}C-fixation. An exception was the cells without nitrogen supply. In these starved cells, ^{14}C-fixation was at its maximum because there was no competition for energy for nitrate uptake (Fig. 7.16).

After one hour of incubation, ^{14}C-fixation values of phytoplankton cells supplied with high nitrate concentrations (10-1000 µg at l^{-1}) began to increase at rates higher than those of cells supplied with 1 µg at l^{-1}. This led, after two hours, to a separation of cells supplied with 1 µg at Nl^{-1} from those supplied with higher concentrations. It appeared that phytoplankton supplied with high nitrate concentrations assimilated enough nitrogen to decrease their starvation state and to direct their energy sources towards ^{14}C-fixation and productivity.

This is emphasized by the increase in ^{14}C-fixation according to the concentration of nitrate at the later stages of the experiment (Fig. 7.16).

Another experiment was conducted to investigate the effect of cell adaptation to different nitrate concentrations before the incubation with ^{14}C. It was found that Thalassiosira cells adapted to nitrate

before incubation have shown the expected relationship between nitrate concentration and ^{14}C-uptake (Fig. 7.17). It can be concluded from the previous two experiments that nitrate-starved cells may give an unexpectedly inverse relationship between ^{14}C-fixation and the nitrate concentration. This is due to competition for energy sources between ^{14}C-uptake and the nitrate uptake. After sufficient nitrate is taken up and the competition stage is over, ^{14}C-fixation increases with the high nitrate concentrations.

Several workers have suggested that electrons are needed to reduce nitrate in the process of nitrate assimilation (Warburg and Negelein, 1920; Van Niel et al., 1953). Thomas et al. (1976) suggested that in nitrogen deficient Chlorella cells, nitrate reduction to nitrite can proceed well by electrons derived from dark reactions but that the nitrite reduction step takes electrons both from dark respiration and from the photochemical mechanism. This means that during competition for energy sources, the nitrate-deficient Thalassiosira and A. japonica cells used the limited energy sources (electrons) for the reduction of nitrite at the expense of ^{14}C-uptake.

The change of chlorophyll a content in Thalassiosira sp. cells supplied with different nitrate concentrations was investigated (Fig. 7.18). Chlorophyll a content in cells supplied with 500 µg at Nl^{-1} increased rapidly which indicates the need for nitrate to retain the normal metabolic processes. In nitrate-starved cells this need for nitrogen was fulfilled at the expense of CO_2-fixation (Fig. 7.16).

Enrichment experiments

a. Variable concentrations of phosphate and nitrate:

Size class 0.45-5 μm:

The pattern of ^{14}C-fixation by cells in this size class was very similar in all the cultures (Fig. 7.19), but there was relatively higher fixation in the cultures enriched with 10 μg at Pl^{-1}, 5 μg at Nl^{-1} and 50 μg at Nl^{-1}. Bearing in mind that the original phosphate concentration in the collected seawater was 0.8 μg at Pl^{-1}, and nitrate concentration was 13.6 μg at Nl^{-1}. This means that in cultures with low ^{14}C-fixation (control, and 0.5 μg at Pl^{-1}) there was a possible phosphate limitation. In all the cultures, the highest ^{14}C-fixation was achieved after two days of incubation with additive nutrients. This could be a result of high growth rate characteristic of small cells (Munk and Riley, 1952; Eppley et al., 1969; Eppley and Thomas, 1969; MacIsaac and Dugdale, 1969; Friebele et al., 1978).

Size class 5-20 μm:

Phytoplankton cells in this size class have shown a strong response to the nutrient enrichment (Fig. 7.20). It seems that the increase in nutrient concentrations has increased nutrient uptake and hence, the growth rate. This can be seen in Figure 7.20, 3, 4 and 5. The highest total of fixed ^{14}C-fixation was found in cultures enriched with 5 and 50 μg at Nl^{-1} (20% and 27% respectively of the total ^{14}C-fixed by phytoplankton of this size class). This may have been the result of an increase in cell number due to the presence of nitrogen which is an essential element in the structure of proteins and amino acids, and an important factor in cell division. In most of the cultures, the highest ^{14}C-fixation was found after two days of enrichment. This may

have been the result of low half-saturation constant and high growth rate (Malone, 1980).

Size class > 20 µm:

^{14}C-fixation in this size class increased slowly, reaching its maximum after four days of incubation (Fig. 7.21). This follows the point of view which argues that large phytoplankton cells have high half-saturation constant and low growth rate (Eppley et al., 1969). On the other hand, the level of fixation was lower than that carried out by the 5-20 µm size fraction which may have been due to low cell number resulting from low growth rate.

In all the previous graphs (Figs. 7.19 - 7.21), high ^{14}C-fixation was of short duration and did not continue for more than four days. This can be seen even in cultures with very high ^{14}C-fixation (Fig. 7.20, 1, 2). It is possible that the growth was eventually limited by phosphate which was present at the start of the experiments in low concentrations (0.8 µg at Pl^{-1}).

b. Variable nitrate concentrations + constant phosphate:

In these experiments, phosphate was added to each culture to give 20 µg at Pl^{-1}, which is much higher than limitation concentration.

Size class 0.45-5 µm:

The ^{14}C-fixation values by this class were very low, except in the culture enriched with 50 µg at Nl^{-1} (Fig. 7.22). In general, the enrichment of sea water with nitrate increased ^{14}C-fixation when compared to the control. The highest fixation value was found in culture with nitrate concentrations similar to those found in Swansea

Bay (see Chapter III). Addition of very high nitrate concentration (500 µg at Nl^{-1}) increased the length of time over which ^{14}C-fixation occurred but the amount of ^{14}C-fixation did not reach such high levels as with the addition of 50 µg at l^{-1}; in the latter case the peak of fixation was obtained within a short period and then dropped off. It can be concluded that nitrate is needed by organisms in the size class 0.45-5 µm up to the level encountered in the natural environment to increase productivity, but very high concentrations may cause an inhibition of ^{14}C-fixation for a short period at least.

Size class 5-20 µm:

When nitrate was added in concentrations similar to those found in Swansea Bay, it stimulated ^{14}C-fixation (Fig. 7.23, 2). But when it was added in very high concentrations (500 µg at l^{-1}), it inhibited the ^{14}C-fixation for a period of time. The fixation was then increased possibly because nitrate concentration dropped to the level (< 50 µg at l^{-1}) found in the natural environment (Fig. 7.23, 3). Very high values of ^{14}C-fixation were found in this size class. This indicates that phytoplankton > 5 - < 20 µm dominated the cultures most of the time and they were more successful competitors than the phytoplankton of the other size fractions.

Size class > 20 µm:

^{14}C-fixation by phytoplankton > 20 µm did not increase to the level reached by phytoplankton between 5-20 µm (Fig. 7.24). Since they both had a similar initial ^{14}C-fixation value prior to the addition of nutrients, the high increase of ^{14}C-fixation by phytoplankton < 20 µm, while the fixation by cells > 20 µm increased very little, supports

the suggestion that small cells have higher growth rates (Eppley et al., 1969). The extended period over which ^{14}C-fixation took place is shown in Figure 7.19, 3 and supports the suggestion that phosphate may have been limiting phytoplankton productivity in the later stages of the experiment conducted with low phosphate concentrations (Figs. 7.19 to 7.21).

c. Variable nitrate concentrations + constant phosphate and silicate: In this experiment phosphate and silicate were added in quantities well above limiting concentrations.

Size class 0.45-5 μm:

There was no significant change in the level and duration of ^{14}C-fixation by this size class except in the culture enriched with 500 μg at Nl^{-1} (Fig. 7.26). This increase occurred after the nitrate concentration dropped to ca. 50% from its original value (Fig. 7.25). This supports the previous findings that exceptionally high nitrate concentrations inhibit ^{14}C-fixation by natural phytoplankton populations.

Size class 5-20 μm:

The addition of nitrate increased ^{14}C-fixation by phytoplankton in the size class 5-20 μm by ca. 40% (Fig. 7.27, 2, 3). Fixation by cells enriched with 500 μg at Nl^{-1} remained at a relatively high level after reaching its maximum value on the sixth day of the experiments. This was probably due to the presence of enough nitrate to sustain the growth of phytoplankton at a relatively high growth rate (Figs. 7.25 and 7.27, 3). The fixation by cells enriched with 50 μg at Nl^{-1},

and those in control dropped heavily on the eighth day, which may have been due to a nitrate decrease to undetectable concentrations on the sixth day of the experiment (Figs. 7.25, 7.27, 1 and 2).

Size class > 20 μm:

The ^{14}C-fixation in this size class was stimulated by the addition of nitrate (Fig. 7.28). Although nitrate did stimulate the increase in ^{14}C-fixation, the duration of this relatively high fixation was different between culture enriched with 50 μg at Nl^{-1} and that enriched with 500 μg at Nl^{-1}. ^{14}C-fixation drop in the former was due to the exhaustion of nitrate, while it was sustained at relatively high levels in the latter because nitrate remained in relatively high concentrations (Fig. 7.25).

It can be concluded from all the previous enrichment experiments that nitrate stimulates the growth of all phytoplankton size fractions at different rates. The rate of carbon fixation and hence productivity was related to phytoplankton cell size and ability to compete. It was found that phytoplankton in the size class 5-20 μm were able to grow and fix ^{14}C at rates higher than those of cells > 20 m. This may have been due to the higher surface area/volume ratio, low half-saturation constant and high growth rate. The importance of this finding for nanoplankton (< 20 μm) is that in Swansea Bay such organisms may dominate the phytoplankton.

CHAPTER VIII

FINAL DISCUSSION AND CONCLUSIONS

Although phytoplankton in Swansea Bay have been studied by many people, there has been no intensive study of their primary productivity. The preliminary objective of the present study was to investigate the net primary productivity of phytoplankton and to relate it to the environmental factors which may have a controlling effect. Another part of the research was to study plankton cell number and phytoplankton biomass. Phytoplankton net with mesh size ca. 50 μm was used. It was preferred to a net with a smaller mesh size. This was because very fine phytoplankton nets may be clogged, especially when we know that Swansea Bay has high turbidity due to suspended particles (Joint, 1980).

Diatoms have been found to dominate the net phytoplankton most of the year. Flagellates occurred for short periods in relatively small numbers (Chapter IV, Figures 2 and 3).

On many occasions no correspondence was found between net phytoplankton number and biomass (chlorophyll a m^{-3}). Examples are October, 1983 (A, B), March, 1984 (B) and June, 1984 (A). See Chapter IV, Figures 4 and 5. The correlation coefficient (r) for phytoplankton biomass and net phytoplankton cell number was + 0.387 (Station A) and + 0.384 (Station B). Paulraj and Hayward (1980), studying phytoplankton at Mumbles Pier (Swansea Bay), found no correlation between net phytoplankton cell number and biomass. These findings suggest that the weak correlation in the present study may have been a result of either phytoplankton spatial heterogeneity, or numbers of small phytoplankton escaping as a result of the relatively large mesh size. If there was spatial heterogeneity, this would result in a weak correlation between chlorophyll a concentration and ^{14}C-fixation, supposing that chlorophyll a concentration represents the biomass of the

total phytoplankton community.

Since chlorophyll a concentration and ^{14}C-fixation were determined using the same water sample from each station, the strong correlation between these two parameters (r [A] = + 0.82, r [B] = + 0.79) means that part of the phytoplankton was missing.

Eppley (1972) suggested that high assimilation numbers (primary productivity/biomass) are associated with small cells. High assimilation numbers have been found on many occasions in the present study (Chapter IV, Table 4.1). Zooplankton grazing is believed to have a minor effect on phytoplankton biomass because zooplankton occurred in relatively small numbers most of the year, except on one occasion (see Chapter IV, Figure 8).

The different phytoplankton species showed different responses to the changing environmental conditions. Individuals of some phytoplankton species have been found to occur in different environmental conditions, e.g. Biddulphia sinensis, Bacillaria paxillifer. The existence of physiological races for B. sinensis and B. paxillifer has been used as an explanation for their occurrence at different times of the year (Tait, 1981). The classification of each species into physiological races was based on their preference for a different temperature range at different times of the year (see Chapter V). Other species occurred mainly at certain times of the year. For example, the highest number of Chaetoceros sp. was found in the spring over a range of temperature from 7-10°C. This range of temperature is thought to be the preference range of Chaetoceros sp. (see Chapter V). Some other species of phytoplankton occurred on one occasion only during the whole period of sampling, e.g. Rhizosolenia delicatula. It occurred in high numbers when the measured environmental conditions were very specific

to that period. Rhizosolenia delicatula occurrence took place in the summer of 1984. At that time, the nutrient concentrations were very low, temperature was high (14-15°C) and salinity was relatively high (30-31‰). From these findings it seems that phytoplankton species succession in Swansea Bay was controlled by more than one factor, but the main factors were temperature and sunshine hours in the winter, and nutrients in the summer (see Chapters III and IV).

From the findings of the growth measurements (cell number, biomass and productivity) and the suggestion that small phytoplankton may have been significant contributors to the total primary productivity, a one-year-long study of primary productivity by phytoplankton size classes was carried out. In the fractionation study, it has been found that there was no obvious seasonal variation in primary productivity by phytoplankton of the size classes 0.45 - 1 μm and 50 - 80 μm. A trend towards seasonal variation has been found in the productivity by phytoplankton > 5 - < 20 μm in diameter. From the data representing the contribution of each size class to the total monthly productivity, it was found that phytoplankton > 5 μm and < 20 μm were the major contributors to productivity during the spring and summer periods. During these periods, the nutrient concentrations reached very low values, and temperature and sunshine hours reached their maximum values (see Chapter III). The occurrence of small phytoplankton cells (> 5-< 20 μm) mainly in periods characterized by low nutrient concentrations, suggests that they have higher uptake rates than larger phytoplankton cells (Eppley et al., 1969). See Chapter VI.

From all the previous findings, it seems that the onset of specific environmental conditions could result not only in the disappearance of a species and the occurrence of another, but even a group

of phytoplankton of certain size class may be replaced by another with better ability to adapt to the new environmental conditions.

On some occasions differences have been found in the phytoplankton populations or total cell numbers between the two stations studied. These differences were very difficult to explain especially when, at the time of the differences, the measured environmental conditions were very similar. However, this would suggest that factors other than the measured nutrients, salinity or temperature may have been responsible for such differences. Lekan and Wilson (1978) found that chlorophyll a patches at wavelengths greater than 20 km were caused by nutrient distribution, while patchiness over wavelengths less than 20 km were related to physical processes. Swansea Bay represents a complex hydrographical system. Several currents occur in this relatively small area. The main current is the rectilinear current parallel to the northern boundaries of the Bristol Channel, with an anticlockwise eddy circulation in the western half of Swansea Bay (Collins et al., 1979). These are accompanied by a very strong tidal current. Joint (1980) studying phytoplankton production in Swansea Bay during August, 1977, found chlorophyll a patches at some stations distributed over a grid located just at the outside boundaries of the bay. He found that the increase in chlorophyll a at certain locations in the period between two cruises is greater than would be expected from the normal increase in chlorophyll a during that period. He suggested that benthic algae derived from the inshore area of the bay may have been responsible for such an increase. The sampling stations of the present study are located at the eastern half of the bay, in an area where the anticlockwise eddy circulation might emerge from the rectilinear current (Ferentinos, 1978). The sampling area in this study is further from

the shore than the area where Joint (1980) found the chlorophyll a patchiness. It is likely that localized physical conditions resulting from the complex hydrodynamics of Swansea Bay may have been responsible for the observed differences in this study between Stations A and B. More studies are needed to investigate this problem.

Several assumptions were made based on the findings of the field work of the present study. Two of these assumptions are particularly important. First, contrary to the results and conclusions reported in previous studies (see Chapter III), nitrate showed a very distinct seasonal variation. Nitrate concentration reached undetectable values in the summer, which suggested that it may have been limiting to the phytoplankton growth in Swansea Bay. Second, from the size fractionation field studies, it has been found that phytoplankton less than 20 μm are major contributors to primary productivity in Swansea Bay. These assumptions were investigated in the laboratory. Special attention was given to the response of phytoplankton (either in prepared media or in natural sea water), according to their cell size, to the changes in the growth conditions.

From the experiments conducted to determine ^{14}C-fixation values by phytoplankton grown in mixed culture, it has been found that smaller cells fixed more carbon because of their high growth rate (Thalassiosira sp.) and high uptake capacity per unit surface area (P. micans). See Chapter VII. Enrichment experiments were conducted to investigate the effect of changing nitrate concentrations on the ^{14}C-fixation by phytoplankton size fractions. Phytoplankton size classes responded differently to nutrient enrichment. When enriched with nitrate, phytoplankton cells > 5 - < 20 μm showed a tremendous increase in ^{14}C-fixation in comparison to 0.45 - 5 μm and > 20 μm size

classes (see Chapter VII).

The importance of nitrate to phytoplankton was further investigated using nitrogen-starved cells of Asterionella japonica and Thalassiosira sp.. When these starved cells were given nitrate, they directed their energy sources towards nitrate uptake at the expense of ^{14}C-fixation. This is because nitrate was urgently needed to maintain their physiological activities. This need was illustrated by the increase in chlorophyll a content of the starved cells when supplied with nitrate (see Chapter VII).

In this study, an attempt was made to investigate the primary productivity of phytoplankton in Swansea Bay and the role of picoplankton and nanoplankton in this productivity. Also, the role of nitrate as a controlling factor to primary productivity received special attention.

From the findings of this study, it can be concluded that nanoplankton play a significant role in the primary productivity of Swansea Bay, and this role is controlled, to a high degree, by the level of nitrate concentration in the sea water.

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